

A STUDY OF CERTAIN EFFECTS
OF THE TEMPERING PROCESS UPON
THE WHEAT PROTEINS.

A thesis submitted to the faculties of
The School of Engineering and the Graduate School
The University of Kansas

For

THE DEGREE OF CHEMICAL ENGINEER

By

MAX C. MARKLEY

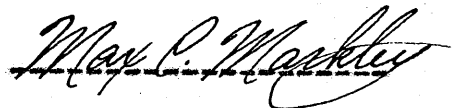
1929

PREFACE

1

This thesis is respectfully submitted to the faculties of the School of Engineering and Architecture and the Graduate School of the University of Kansas in partial fulfillment of the requirements for the degree of Chemical Engineer.

This thesis represents the results of a research upon a phase of the flour milling process, and was carried out by the author in the laboratory of the Slater Mill and Elevator Company. The author is indebted to this company for materials and equipment used in this research.

A handwritten signature in cursive script, reading "May C. Mackley". The signature is written in dark ink and is positioned above a horizontal line.

Slater, Mo.

May 3, 1929.

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Until about forty years ago, grain was conditioned before grinding only by dry cleaning methods, using chiefly screens and air suction. At the present time the dry cleaning methods are much improved, so that now an almost perfect separation of the grain from all foreign matter and dirt is effected. There are also wet cleaning systems in use which wash the wheat or other grain free from dirt and even smut infections.

There are today a number of processes in general use which change the physical and chemical nature of the grain in preparation for the milling process. The most common of these tempering methods is the addition of a small quantity of water to the grain, agitating it to obtain a uniform mixture, and then allowing the grain to lie in a bin for a period of time varying from a few minutes to a week or more. This wetting of the grain, accompanied by the subsequent rest period, may be done once, twice, or even three times.

Dry heat is frequently used. The temperature of the wheat is rarely raised more than thirty or forty degrees. The grain is sometimes heated several times before it is milled. Live steam is often used, especially just before the wheat goes to the first break rolls.

These tempering methods have a profound influence upon the physical nature of the wheat. The berry swells and becomes softer. The bran coating becomes tougher, so that the break rolls lay it out in large flakes instead of in small chips, as is the case when dry wheat is ground. The white internal fiber of the endosperm is toughened so that the subsequent grinding of the middlings does not pulverize it, and it can be bolted out of the finished flour.

Along with these physical changes there are probably chemical changes, but little research work has been done on this problem. This investigation was therefore instigated to determine if there were any such chemical changes, and if so, to measure the extent of them, and establish the general laws of their occurrence.

A survey of the available literature showed no work bearing directly upon the subject, but there was a wealth of data upon allied subjects, which became of interest as the investigation progressed, aiding in interpreting the results.

At the beginning of this work it was decided to confine present investigations to the protein changes. The results of many investigators have shown that wheat proteins are made up of a number of true proteins and a small quantity of protein degradation products. The principal proteins are a prolamin, gliadin (Osborne and Voorhees, 1893); a glutelin, glutennin (Osborne and Voorhees, 1893); an unnamed globulin (Osborne and Voorhees,

1893, Denis, 1859); and an albumin, leucosin (Osborne and Voorhees, 1893, Osborne and Campbell, 1900). There are also small quantities of proteoses and peptides, simple amino acids, and even some ammonia present.

The time-honored method for the study of the wheat proteins has been to separate the individual proteins by their varying solubility in different solvents. Leucosin has customarily been separated by dissolving it out in water. But the work of Johnson and Bailey, 1924, has established that the amount of water soluble nitrogen in a flour suspension in water was determined almost entirely by the hydrogen ion concentration of the suspension. This work casts a reflection upon the custom of calling the water soluble fraction of the wheat protein, leucosin. This relationship was investigated during the course of this work.

The proteins soluble in dilute salt solutions have ordinarily been classed as globulin. However the recent work of Gortner, Hoffman, and Sinclair, 1929, has shown that individual samples of flour differ markedly in their solubility in solutions of several salts of varying strengths. This investigation makes it improbable that any one salt solution extracts completely any one definite protein with no admixtures of other proteins.

Tague, 1925, has shown that gliadin is appreciably soluble in water, dilute acids, alkalis, salt solutions,

and in methyl alcohol as well as in the regular seventy per-cent ethyl alcohol solution.

After studying the above researches, it was decided that a separation of the proteins by their solubilities would only give misleading and uncertain results. So attention was turned to the simpler nitrogen compounds of the wheat. These can be determined with a high degree of accuracy. Some seven methods of determining these simpler products were discussed by Cairns and Bailey, 1928. For the purposes of this work, the amino-acid content was determined by Sorensen's formol titration method, and non-protein nitrogen by Ritthausen's method as modified by Blish, 1918, and by Olsen and Bailey, 1925. The Ritthausen method precipitates everything down to the peptide and amino-acid stage. A large number of investigators have used these and similar methods to follow the protein changes of wheat under different conditions.

A study of the literature reveals much evidence that germinating wheat is more active in protein splitting enzymes than is dormant wheat. Ballard, 1884, found that germinated wheat had the power of liquefying gluten. Abderhalden and Schittenhelm, 1906, demonstrated the juice pressed from germinating wheat was capable of hydrolyzing polypeptides. Vines, 1906, 1909, established the presence of proteases in both germinated and ungerminated wheat.

Sherwood and Bailey (1926) show large increases in the simpler nitrogen compounds during the germination of wheat.

Kedzie (1893), Teller (1898), Nedokutschajew (1902), Brenchley and Hall (1908-10), Thatcher (1915), Swanson, Fitz, and Dunton (1916), Sharp (1925), Woodman and Engeldow (1924), and Mangels and Stoa (1928), all show that the simpler nitrogen forms tend to disappear as the wheat ripens. Eckerson (1917) came to the following conclusions:

"The nitrogen compounds, aside from the aleurone and protoplasm, in the endosperm just before ripening of the grain are: much asparagine, considerable arginine, histidine, and some leucine. No glutamine was found.

"On desiccation of the grain, protein appears in the storage cells; the amino-acids and most of the asparagine disappear. The protein has the physical characters of gluten.

"Formation of the storage protein in wheat seems to be a condensation process, and it takes place on desiccation of the wheat kernel."

Olson (1917) found that the higher the temperature at which immature harvested wheat was dried, the higher the amide-nitrogen content of the resulting flour; but if the sample of immature wheat were allowed to stand in the laboratory before subjecting to heat, the resulting flour would have a lower amino-nitrogen content.

Sharp (1925) found that the amino-nitrogen content of ripening wheat decreases until the moisture content is reduced to thirty-five per-cent, and thereafter remains constant.

m In main, the official methods of the American Association of Cereal Chemists were used, with only minor variations, which did not affect the accuracy of the work. All results except moisture were reported on a fifteen per-cent moisture basis.

a. Moisture.

The samples of wheat when dry enough were ground on a laboratory burr mill. Two grams of the ground material were rapidly weighed into a freshly dried and tared aluminum moisture dish with cover. The dish was then placed in a Freas air oven at one hundred thirty degrees centigrade for one hour; then withdrawn, covered at once, and placed in a desiccator over sulphuric acid to cool. It was weighed back as soon as cool, and the loss in weight considered as moisture. If the sample was too wet to grind, the entire wheat was used, and the drying continued to constant weight, usually about three hours.

b. Total Nitrogen.

The Gunning modification of the Kjeldahl method was used. It consists in weighing one gram of the ground sample into an eight hundred cubic centimeter long-necked Kjeldahl flask. Fifteen cubic centimeters of concentrated sulphuric acid, seven grams of sodium sulfate, and one-tenth gram of copper sulfate were added, and the flask was heated on a high heat electric burner until the contents were a clear blue color and reduced to a volume of ten

cubic centimeters. The flask was then cooled, and the contents made up to approximately five hundred cubic centimeters with water. An excess of caustic soda was added and the five hundred cubic centimeters distilled through a block tin still into twenty cubic centimeters of tenth normal sulphuric acid. The remainder of the acid was titrated with tenth normal sodium hydroxide. A blank was run on the reagents.

c. Preparation of Wheat Extract.

Forty grams of the finely ground wheat meal were weighed accurately and placed in a one-liter erlenmeyer flask with four hundred cubic centimeters of distilled water saturated with toluene. The contents were thoroughly mixed and the flask placed in an air thermostat at thirty degrees centigrade for two hours, being shaken every fifteen minutes. At the end of two hours the supernatant liquid was decanted through a coarse paper filter. If the sample was too wet to grind on the burr mill, forty grams of the whole wheat were weighed quickly and thrown into a mortar. The wheat was then ground with successive small portions of the four hundred cubic centimeters of water until well disintegrated.

d. Amino Acids by Sorensen's Method.

A one hundred cubic centimeter aliquot of the water extract of the wheat was titrated with tenth normal sodium hydroxide solution using phenolphthalein as indicator.

Then to the neutral solution, twenty cubic centimeters of freshly neutralized formaldehyde were added, and allowed to stand fifteen minutes. At the end of fifteen minutes it was again titrated with tenth normal sodium hydroxide. This second titration represents the amino-acid content of the sample. A figure representative of the amino-nitrogen content can be had by multiplying the amino-acid titration by fourteen-hundredths, giving the amino-nitrogen in per-cent.

Denham and Blair (1927) claim the time of standing in the thermostat causes higher results because of proteolytic activity, and suggest several determinations with varying times, and then plotting percentage against log time to get a curve which can be interpolated back to the initial time of sampling. However, as their curve is open to criticism, in that it is a curve and not a straight line as they assert, and also as the two-hour time was a constant throughout the series, it was decided to ignore their criticism.

The initial titration in this determination was used as the measure of the titratable acidity of the wheat.

c. Water Soluble Nitrogen.

A fifty cubic centimeter aliquot of the wheat extract was placed in a kjeldahl flask and the nitrogen determined in the same manner as the total nitrogen.

In order to prevent foaming, the sodium sulfate was not added until the water was evaporated and the heavy white acid fumes appeared.

f. Copper Non-precipitable Nitrogen.

A fifty cubic centimeter aliquot of the wheat extract was placed in a one hundred cubic centimeter graduated flask. A few drops of phenolphthalein were added, and then twenty-two cubic centimeters of tenth normal sodium hydroxide were run in from a burette. Copper sulfate solution was slowly added until the solution changed from red to purple, and finally to green. It was then made up to mark with water. After the heavy precipitate had settled, a fifty cubic centimeter aliquot was taken, and determined for nitrogen in the same manner as the water soluble nitrogen. Only in this case no additional copper was added.

Experimental Method.

The method of tempering adopted for this work closely paralleled the standard tempering practice. Unless otherwise noted in the discussion, the following method was used: Two hundred grams of the sample of wheat were weighed into a pint Mason fruit jar, and twenty cubic centimeters of water added. The lid was screwed on without a rubber, and the sample was thoroughly shaken for several minutes until the water was well distributed. The jars were then

set in an air thermostat controlled by a bi-metallic electric regulator. At the end of the respective times, portions of the tempered wheat were withdrawn, and the jars returned to the thermostat.

Variable Moisture Content During Tempering.

A fairly stable sample, number eighteen, of hard winter wheat, was chosen for this series. It was analyzed for moisture, total nitrogen, water soluble nitrogen, copper non-precipitable nitrogen, amino-acids, and titratable acidity. Another portion of the sample was heated forty minutes at one hundred thirty degrees centigrade, and then ground and analyzed.

The amino-acid and water soluble nitrogen showed slight decreases, while the copper non-precipitable nitrogen increased slightly. But on the whole the results varied so little that the differences are within the experimental error, and are probably without significance.

One portion of the sample was tempered with ten per-cent distilled water in the regular manner. This was enough to raise the moisture nearly eight per-cent. Another portion was tempered with sixty per-cent water, which was a slight excess over the maximum water absorption of the wheat. At the end of twenty-one hours, samples of each were withdrawn and heated for forty minutes. They were dry ground along with a sample of the regular temper that had not been heated, and were then analyzed.

The amino-acid content showed a small but marked increase in the regular-temper-no-heat sample. It was slightly higher in the sample tempered with excess moisture

and oven dried before grinding, and decidedly lower in the regular-temper-oven-dried sample. The copper non-precipitable nitrogen showed the same variations as did the amino-acids. The water soluble nitrogen showed an increase in the regular-temper-no-heat sample, but a decrease when this regular-temper sample was heated, and a much larger decrease in the excess-moisture-oven-dried sample.

At the end of twentieth-six hours a portion of each tempered sample was withdrawn. The excess moisture sample was spread out in an open pan and placed in the thermostat at thirty degrees centigrade. The portion of the normal-temper sample was placed in another mason jar, tempered with excess distilled water, and placed in the thermostat.

At fifty-two hours a portion of the original excess-moisture sample was withdrawn, ground wet in a mortar, and analyzed. There was a very great increase in the amino-acids, water soluble nitrogen, and copper non-precipitable nitrogen. Moisture was determined by drying the whole grain to constant weight at one hundred thirty degrees centigrade. The wheat by this time had sprouts a quarter of an inch long.

At the end of ninety-four hours portions of the two original tempers were dried and dry ground. A portion of the regular temper was ground without heating.

At one hundred hours, portions of each of the excess moisture samples were wet ground without heat, and the sample of wet wheat that had been spread out to dry in the open pan was dry ground without heat.

The amino-acid content of the normal tempered sample showed a decrease from the value at twenty-two hours. With the same sample heated for forty minutes before grinding, the amino-acid content was much lower than the normal tempered sample. The sample sprouted and then dried before grinding showed a large increase. This increase was in direct ratio to the time tempered. The sample sprouted then wet ground had an excessive increase which was directly in line with the fifty-two hour analysis. The sample tempered regularly twenty-six hours, then excessively the rest of the time, and wet ground without heat, paralleled the sample tempered all the time excessively. The sample spread out to dry in the pan was very dry when ground, and had an amino-acid content between the regular sample and the excess-moisture-dried sample.

The copper non-precipitable nitrogen and water soluble nitrogen contents followed the amino-acids very closely.

The free amino-acid content of this wheat increased directly with the time, when there was an excess of moisture present at thirty degrees centigrade and no

appreciable desiccation. When desiccation set in, the free amino-acid content decreased directly with the length of time, moisture content, and temperature. At thirty degrees the rate of decrease of the free amino-acids during desiccation was much less than the rate of increase during germination at the same temperature.

When water was added in insufficient quantity to maintain germination, the initial rate of change of the amino-acid content was a component of the rate of increase during germination and the rate of decrease during desiccation. In this sample the germination had entirely ceased by the end of twenty hours.

When heated to one hundred thirty degrees centigrade there was a very rapid decrease in amino-acid content. This decrease was directly proportional to the moisture content. At forty per-cent moisture there was a fifty per-cent decrease in the one hundred-hour tempered sample. But at nine per-cent moisture the decrease was only about six per-cent.

The rates of change of the free amino-acid content of wheat during desiccation and germination may be described as being linear functions of the time, temperature, and moisture content. The reaction probably follows the peptide condensation as worked out by Emil Fischer.

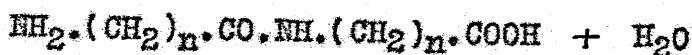


TABLE 1

Variable Moisture.

Treatment	Time Temp. Hrs.	Moisture %	Acidity c.c. H ₂ SO ₄	Amino-N %	Water Sol.N %	Cu non pp.N %
Dry Wheat No Heat	0	9.06	3.33	.0177	.316	.0840
Dry Wheat Heated	0	7.22	1.75	.0167	.286	.0872
Regular Temper No Heat	22	16.86	3.07	.0193	.337	.0890
"	94	16.37	2.6	.0171	.303	.0828
Regular Temper Heated	21	14.51	2.2	.0139	.251	.0810
"	92	11.87	1.78	.0121	.217	.0864
Excess Moisture No Heat	52	40.32	5.7	.0418	.718	.310
"	100	46.65	8.75	.0632	.900	.492
Excess Moisture Heated	22	32.98	3.05	.0213	.204	.0854
"	94	22.88	4.4	.0309	.370	.1548
Regular Temper Excess Moisture	26 74 100	46.82	7.2	.0560	.875	.3680
Excess Moisture Air-dried	26 74 100	8.90	4.53	.0261	.373	.1204

All results except moisture are calculated to 15% moisture basis.

Variable Moisture

Amino-Nitrogen - Tempering Time

% Amino N.

.060

.055

.050

.045

.040

.035

.030

.025

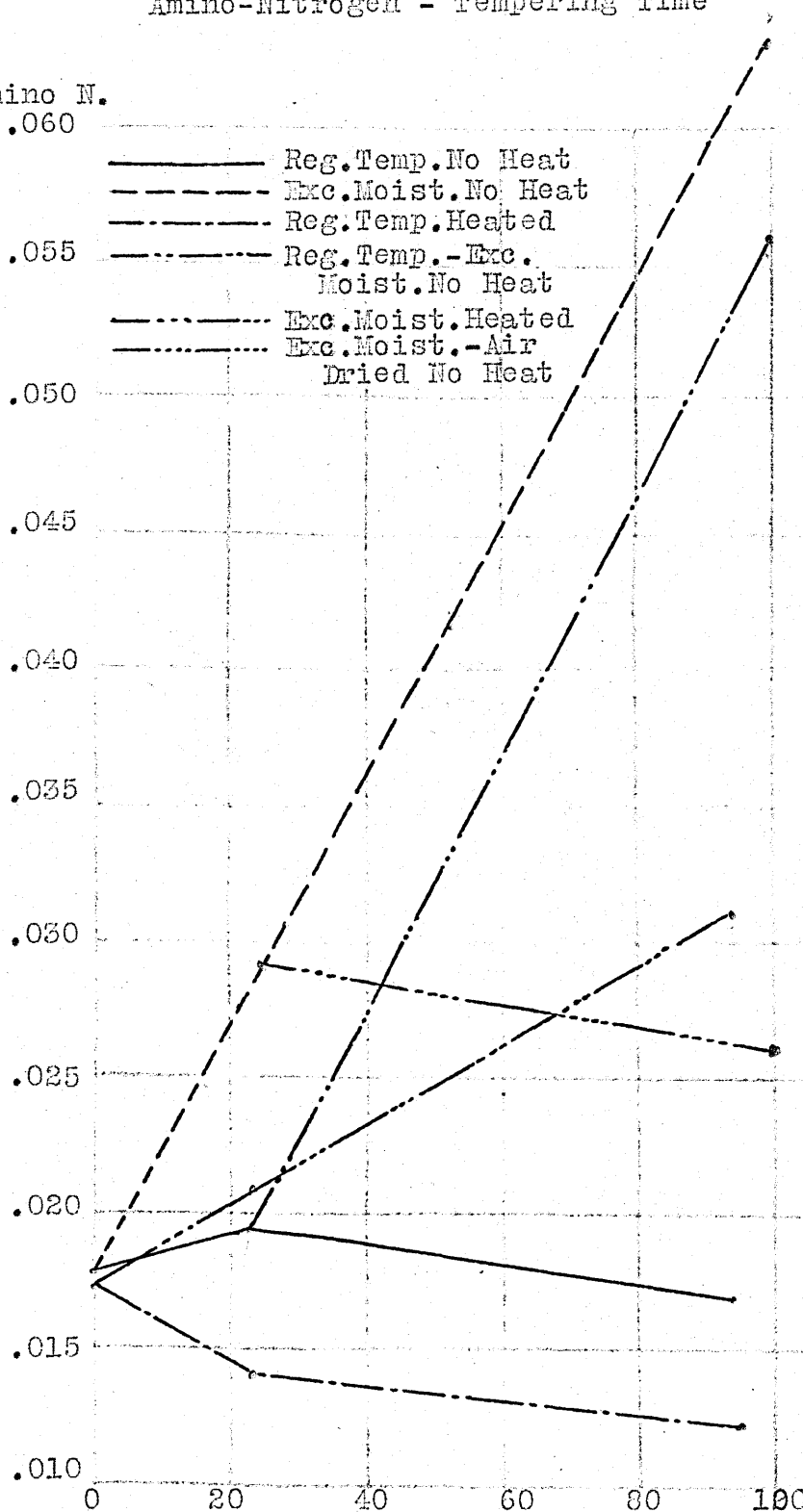
.020

.015

.010

- Reg.Temp.No Heat
- - - Exc.Moist.No Heat
- · - Reg.Temp.Heated
- - - Reg.Temp.-Exc.
- · - Exc.Moist.Heated
- · - Exc.Moist.-Air
- - - Dried No Heat

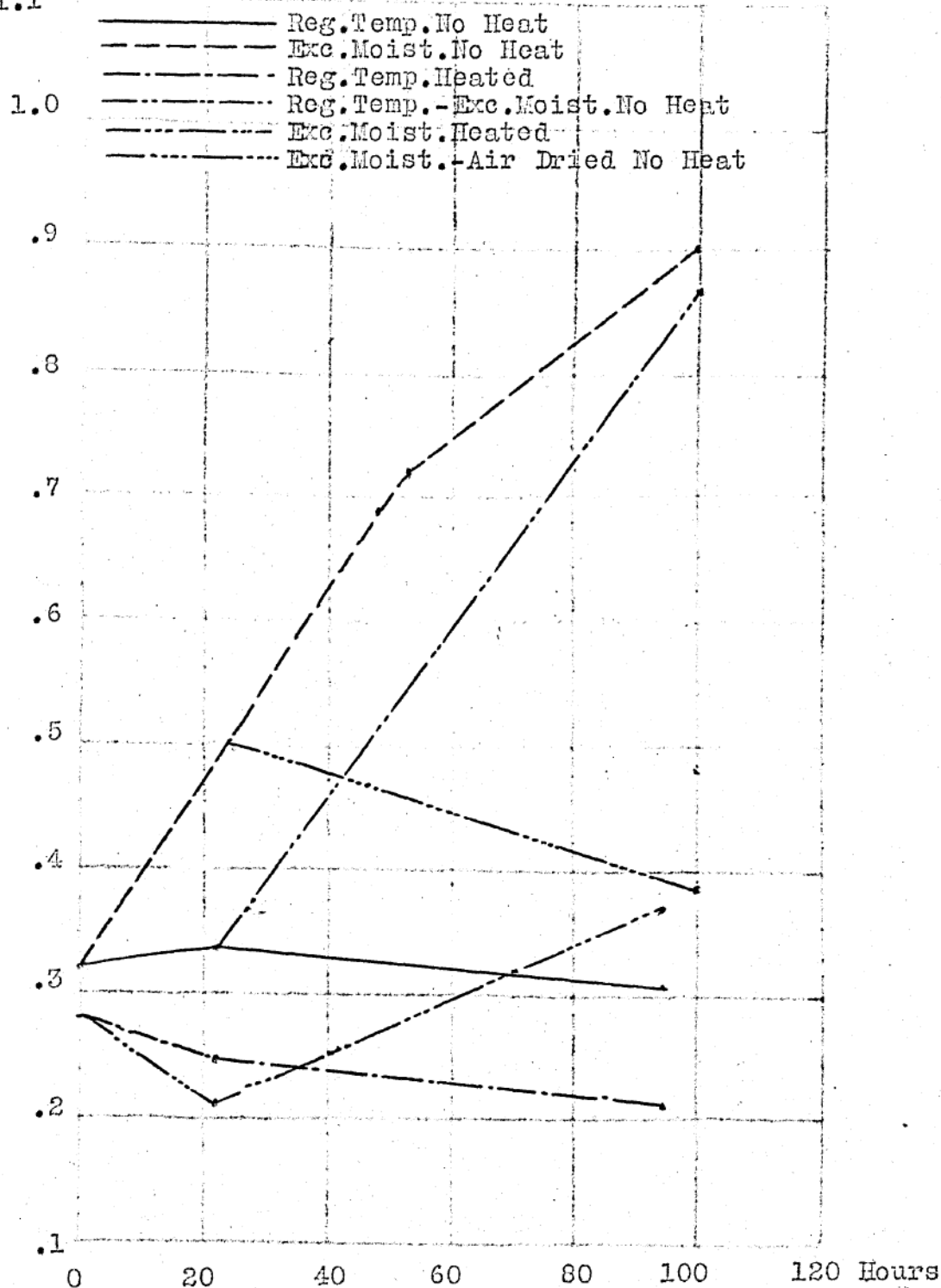
0 20 40 60 80 100 120 Hours



Variable Moisture

Water Soluble Nitrogen - Tempering Time

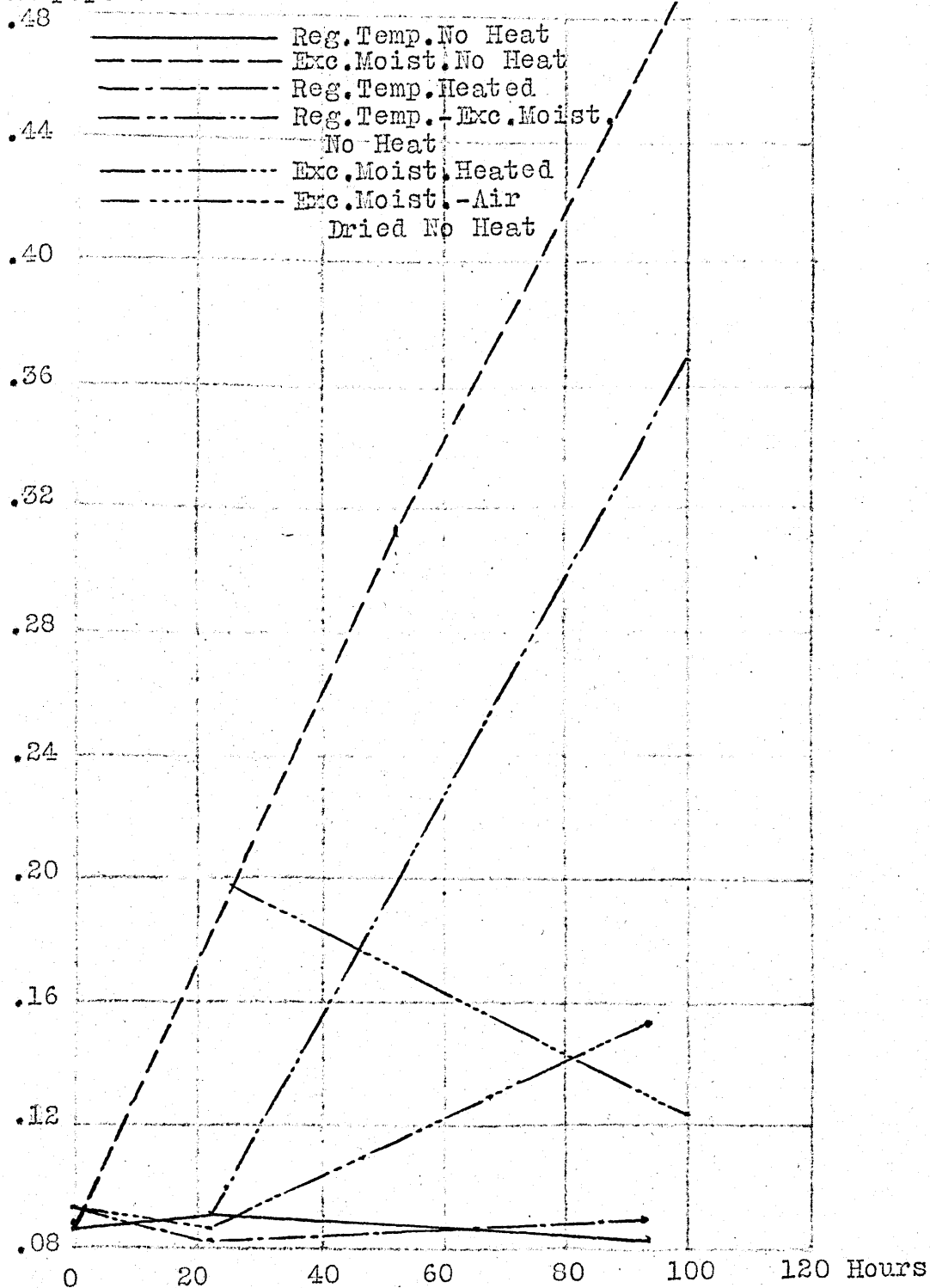
% Water Sol.N.



Variable Moisture

Copper Non-precipitable Nitrogen - Tempering Time.

% Cu non-p.p.N.



Water Soluble Nitrogen - Titratable Acidity

Variable Moisture

%Water Sol. N.

1.1

1.0

.9

.8

.7

.6

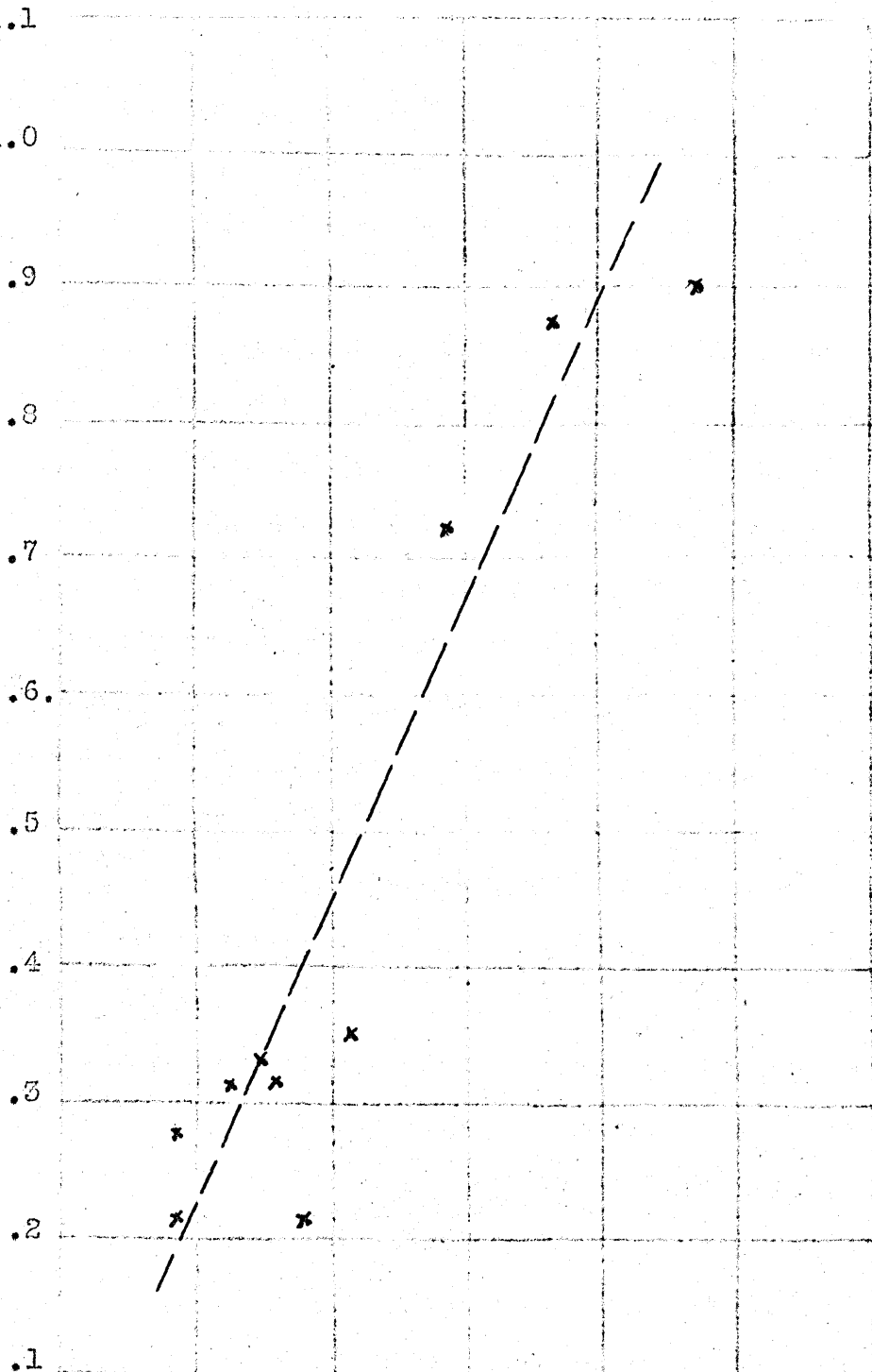
.5

.4

.3

.2

.1

0 2 4 6 8 10 12 c.c.
N/10 H₂SO₄

EFFECT OF VARIABLE TEMPERATURES UPON WHEAT PROTEINS DURING TEMPERING

Sample Number Sixteen was selected for this work. This was a sample of low protein hard winter wheat of the 1928 crop grown at Lewis, Kansas, a station situated on a sandy upland south of the Arkansas River in southwestern Kansas.

This sample was analyzed for moisture, total nitrogen, water soluble nitrogen, copper non-precipitable nitrogen, and amino-acids. Two two hundred gram portions were placed in pint Mason fruit jars, and the lids screwed on without rubbers. One jar was placed in a thermostat at thirty degrees centigrade, and the other was placed outside the north window on the third story of the mill building, at an average temperature of minus eighteen degrees centigrade. The samples were allowed to come to equilibrium with their surroundings, and then were tempered with ten per cent of distilled water, and returned to their respective positions.

At the end of seventeen hours' tempering, one third of each sample was withdrawn, placed in another pint Mason jar, and set in the reverse temperature condition.

At the end of twenty-two hours, one third of each of the two original samples was withdrawn, ground, and analyzed. The wheat held at thirty degrees gave the characteristic soft, flaky grind of well-tempered wheat, while the

frozen sample gave a sharp, granular, yellow chop with no broad bran.

The moisture content of the frozen sample was about one per-cent higher than the sample held at thirty degrees. The frozen sample showed no significant change in water soluble nitrogen, while the sample held at thirty degrees showed a slight decrease.

The amino-acid content likewise showed very little change in the frozen sample, but a very decided decrease in the sample held at thirty degrees. There was a marked decrease in the copper non-precipitable nitrogen content of both samples. However it was the frozen sample that in this case showed the larger decrease.

These results indicate that desiccation had set in from the start with this sample, the particular condensation reaction, however, being a function of the temperature. At thirty degrees the principal reaction is probably the usual peptide condensation. But at temperatures below the freezing point of water the reaction is probably chiefly between the amide derivatives of the simple amino-acids. This reaction, however, has not been studied enough to make any definite statements.

At the end of ninety-four hours* tempering time, the sample held at thirty degrees the entire period, and the sample held at minus eighteen degrees for seventeen hours, then at thirty degrees for seventy-seven hours,

were analyzed. The moisture content of the sample held at thirty degrees the entire time had risen about one percent, while the other sample showed a slight decrease.

The amino-acid content of the sample held the entire time at thirty degrees was the same as at twenty-two hours, indicating that the peptide condensation had stopped. The sample that had first been frozen and then held at thirty degrees showed a marked decrease in amino-acids at ninety-four hours. The amino-acid content was rapidly approaching the minimum value for thirty degrees.

The two frozen samples were analyzed at the end of one hundred-fourteen hours. The sample held at minus eighteen degrees the entire time showed no great change in amino-acid content, only a slight decrease. The sample held seventeen hours at thirty degrees and the rest of the one hundred-fourteen hours at minus eighteen degrees showed an increasing amino-acid content, with a value at one hundred-fourteen hours practically the same as in the dry sample.

Thus it appears that when this sample of wheat was held at temperatures below the freezing point of water, the amino-acid content tended to remain the same, regardless of the moisture content of the wheat. But when the temperature was above freezing, this sample showed a decrease in amino-acid with increasing water content, provided that the water content was kept below that necessary to start and maintain germination.

The water soluble nitrogen content showed a general decrease during the long tempering period with no apparent relationship to temperature. The water soluble nitrogen was closely correlated with the titratable acidity in the tempered samples.

At ninety-four hours, the sample held at thirty degrees the entire period showed an increase in copper non-precipitable nitrogen, while the sample held at minus eighteen degrees for seventeen hours and at thirty degrees the rest of the time had a slight decrease. The two frozen samples both showed a decided decrease in the latter part of the period. The increase in the one sample was probably caused by a secondary reaction setting in late in the tempering period, as that sample showed an unexplained increase in moisture. This secondary reaction may be a splitting of the higher molecular weight polypeptides. This is only a conjecture, and has not been verified.

The increase in moisture content of the frozen samples during the last of the tempering period was due to a heavy snow which covered the loosely closed Mason jars.

TABLE 2

Variable Temperature

Temperature of Tempering °C	Time Temp. Hrs.	Moisture %	Acidity c.c. H ₂ SO ₄	Amino-N %	Water Sol.N %	Cu Non- pp. N. %
Dry Wheat	0	10.28	2.05	.0189	.318	.0850
-18	21	17.07	3.0	.0187	.320	.0712
-18	114	18.50	2.15	.0175	.250	.0528
30	21	15.97	2.95	.0138	.307	.0768
30	92	18.44	2.4	.0138	.272	.0848
-18	17					
30	75					
	92	16.71	2.2	.0157	.266	.0654
30	17					
-18	97					
	114	20.20	2.6	.0186	.287	.0682

All results except moisture are calculated to 15% moisture Basis.

Variable Temperature

Amino-Nitrogen - Tempering Time

%Amino-N.

.022

.021

.020

.019

.018

.017

.016

.015

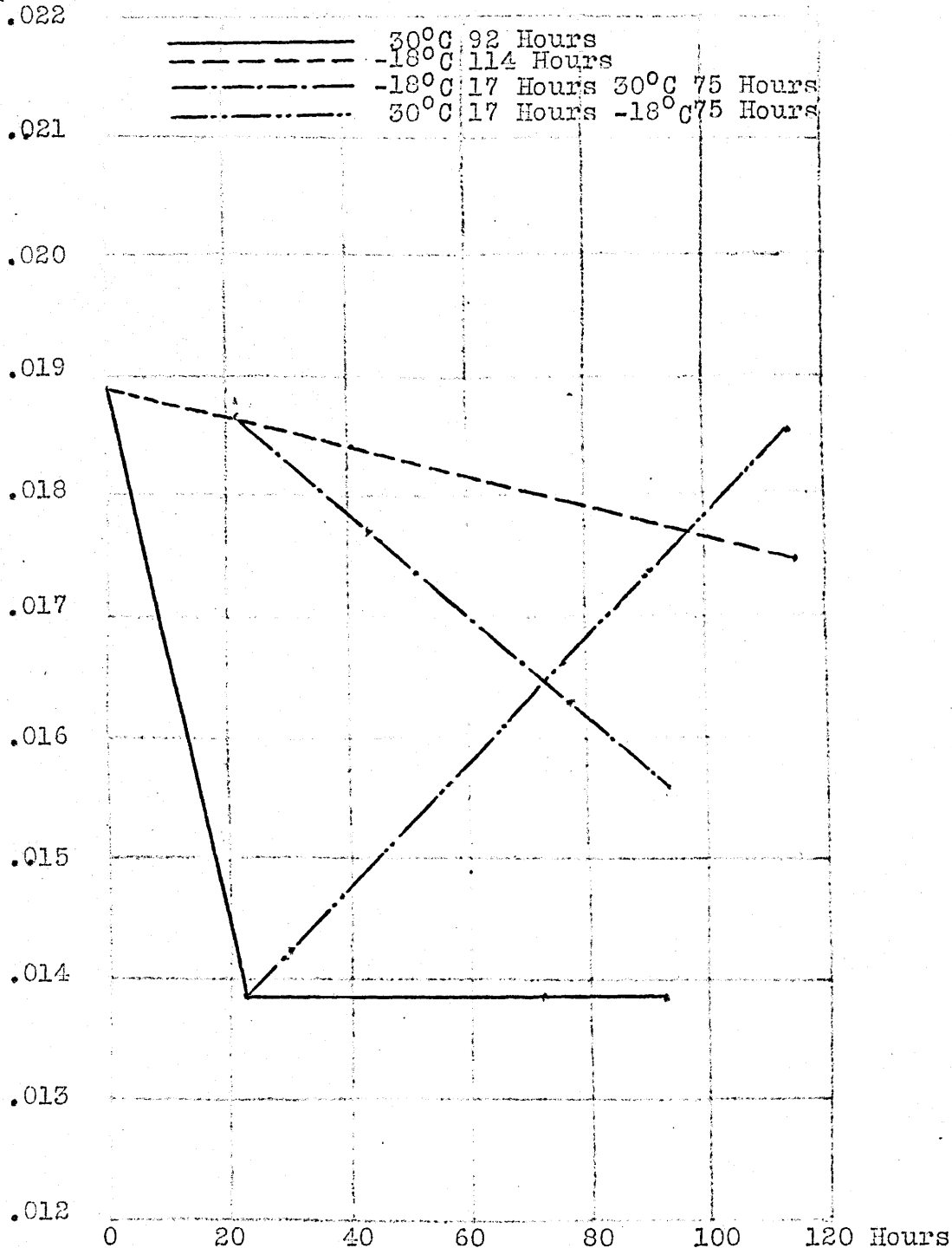
.014

.013

.012

—————	30°C	92 Hours
- - - - -	-18°C	114 Hours
-	-18°C	17 Hours
-	30°C	17 Hours
	30°C	75 Hours
	-18°C	75 Hours

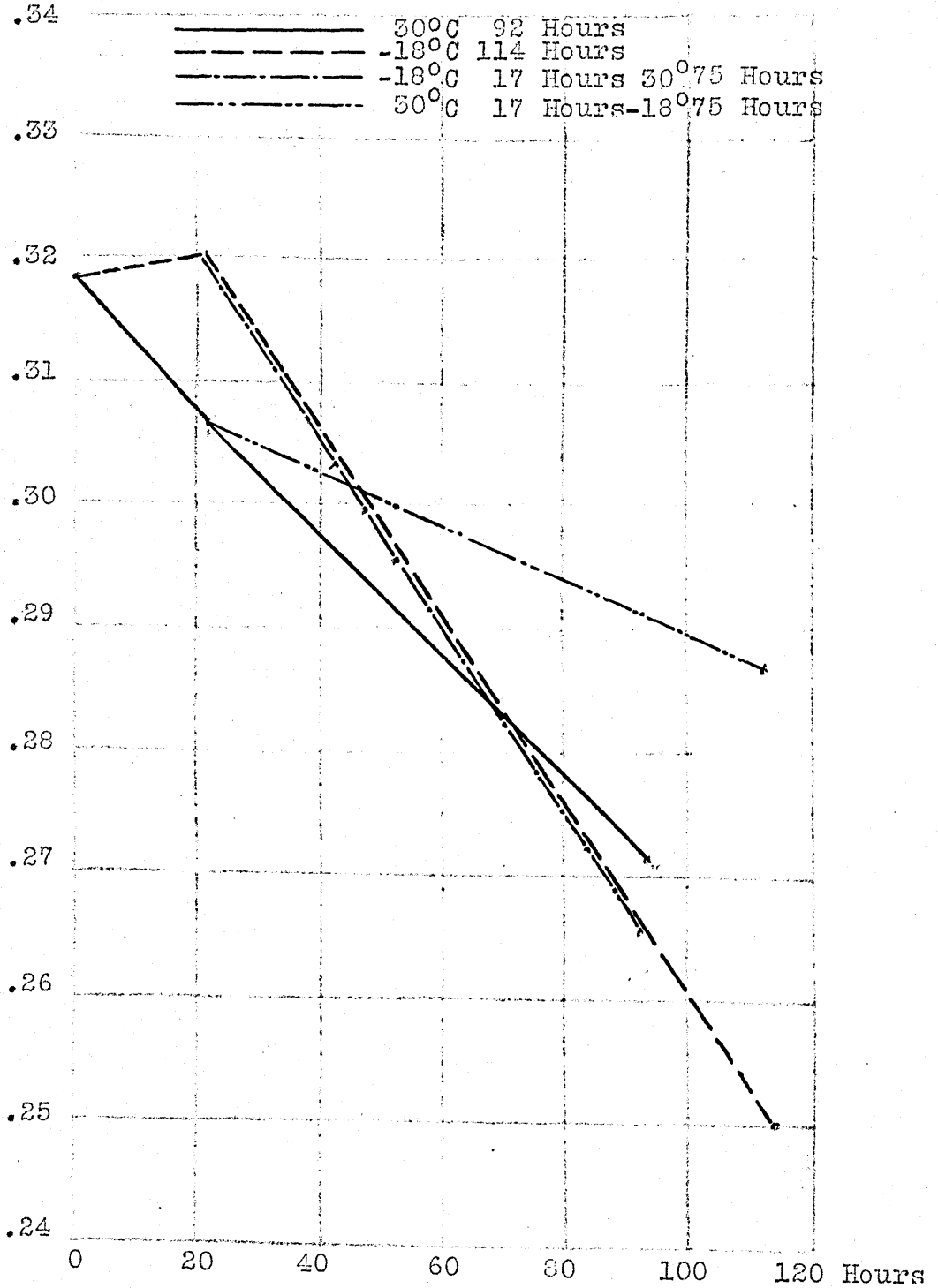
0 20 40 60 80 100 120 Hours



Variable Temperature

Water Soluble Nitrogen - Tempering Time

% Water Sol.N.



Copper Non-precipitable Nitrogen - Tempering Water.

Variable Moisture

%Cu non-p.p.N.

.092

.088

.084

.080

.076

.072

.068

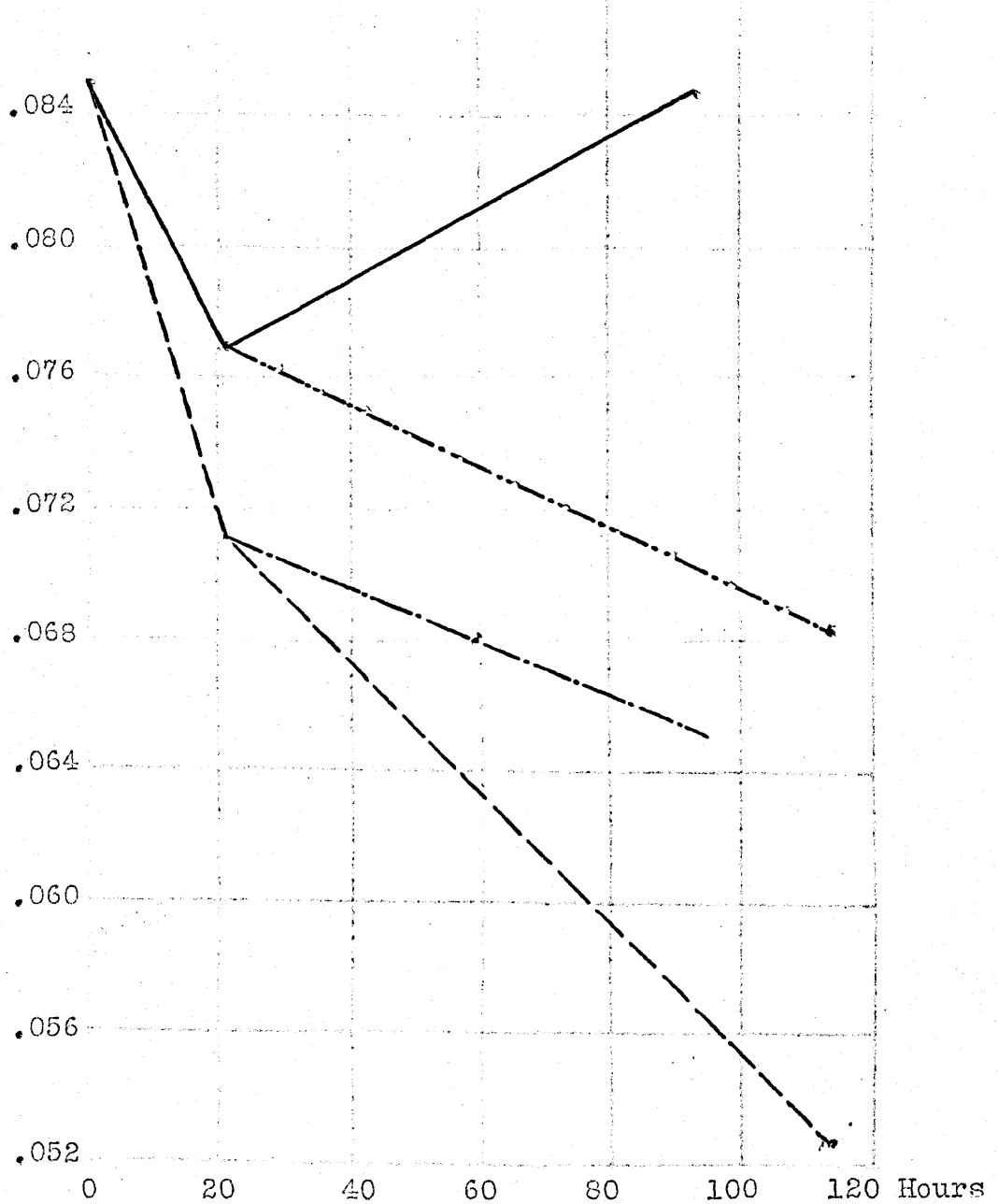
.064

.060

.056

.052

—	30°C	92 Hours	
- - -	-18°C	114 Hours	
- · - · -	-18°C	17 Hours	30°C 75 Hours
- · - - -	30°C	17 Hours	-18°C 75 Hours



Variable Temperature

Water Soluble Nitrogen - Titratable Acidity

% Water Sol.N.
.34

.33

.32

.31

.30

.29

.28

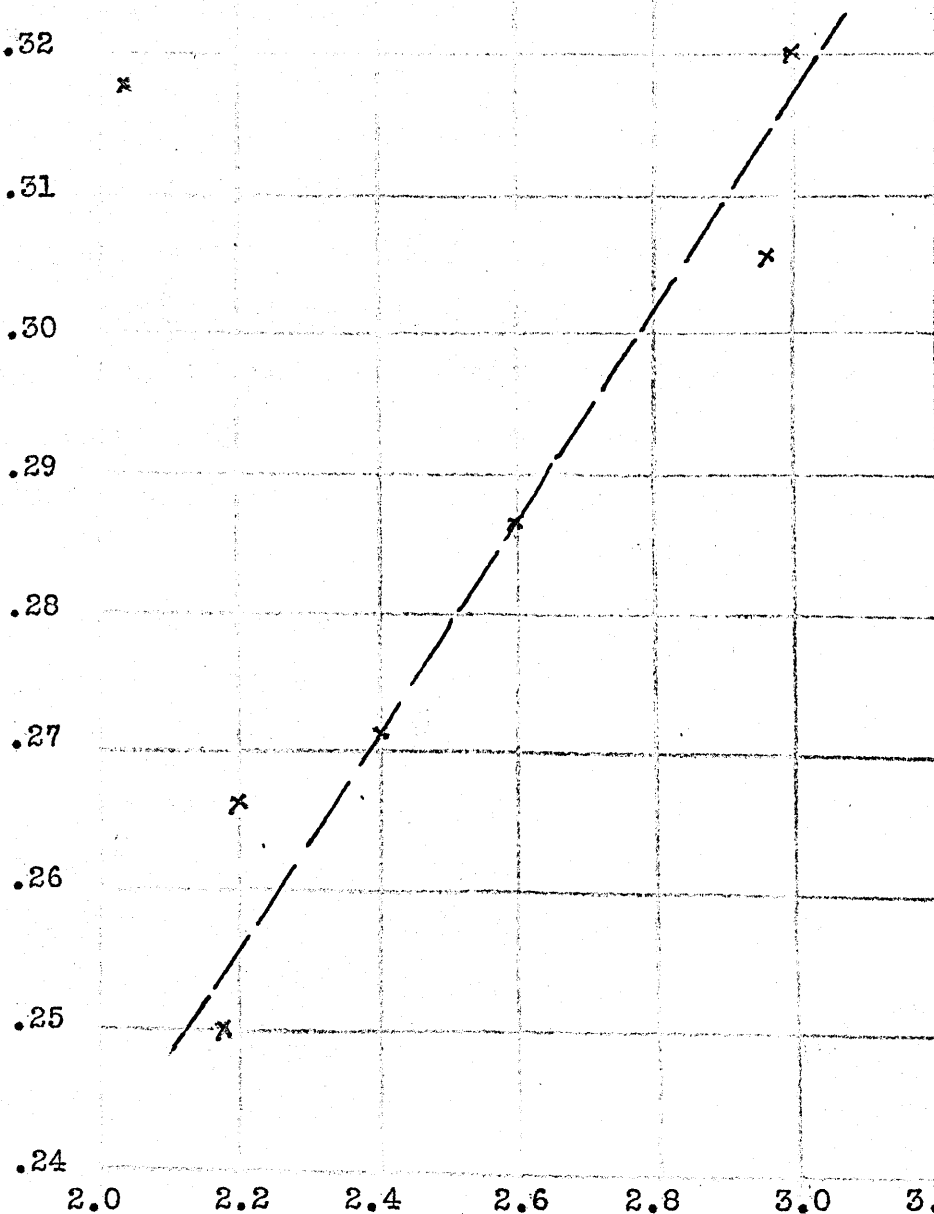
.27

.26

.25

.24

2.0 2.2 2.4 2.6 2.8 3.0 3.2 c.c.
N/10 H₂SO₄



In order to determine whether the different types of wheat have different chemical properties during tempering, or if the differences are environmental, some fifteen samples of wheat were obtained through commercial sources. These samples were so selected as to show both varietal and environmental differences.

The soft red wheats were all grown in a small section of central Missouri, but on three types of soils - high black prairie, clay hill, and sandy river bottom.

The hard red winter wheats were from southwestern Kansas, taking in sandy bottom, sandy prairie, and hard prairie soils. As all were grown close together, climatic conditions should be approximately the same.

Only two samples of Hybrid 128, a white club wheat, were available. One was from the Willamette Valley of western Oregon, and the other was from Umatilla County in northeastern Oregon.

A sample of Soft Federation, a common white wheat, from the same station in the Willamette Valley, and another from the same station in Umatilla County, were secured. Another Umatilla County sample was also studied.

Two samples of Dicklow, another common white wheat, were tempered. One of these samples was from an Idaho country-run car, and the other from an Ogden, Utah, elevator blend.

Description of the Wheat Samples.

No.	Kind of Wheat	Origin	% Moisture	% Total Nitrogen
1	Hybrid 128	Fulton, Oregon	9.10	1.23
2	" "	Vansycle, Oregon	10.47	1.30
3	Soft Federation	Fulton, Oregon	8.94	1.43
4	" "	Helix, Oregon	9.30	1.67
6	" "	Vansycle, Oregon	9.71	1.84
10	Hard Red Winter	Garfield, Kansas	10.78	1.87
11	" " "	Greensburg, Kansas	9.59	1.84
12	Dicklow	Ogden, Utah	10.69	1.71
13	"	Buhl, Idaho	10.36	1.76
14	Soft Red Winter	Slater, Missouri	9.14	1.89
15	" " "	Glasgow, Missouri	10.88	2.01
16	Hard Red Winter	Lewis, Kansas	10.28	1.79
17	Soft Red Winter	Glasgow, Missouri	15.24	1.81
18	Hard Red Winter	Centerview, Kansas	9.06	1.86
19	Soft Red Winter	Glasgow and	10.63	1.95

Carrollton, Missouri

The total nitrogen is calculated to fifteen percent moisture basis.

Four samples of Missouri soft red winter wheat were tempered and analyzed at different stages. Sample Number Fourteen was from a very choice lot of seed wheat grown on a high black prairie near Slater, Missouri. Sample Fifteen was from a lot of wheat grown upon the clay hills near Glasgow. This sample carried the highest nitrogen content of any used in this work. It also had a few sprouted and damaged kernels. Sample Seventeen was grown on the sandy Missouri River bottom near Glasgow. This sample had been carelessly harvested and stored, so when received was about thirty per-cent sprouted, and was slightly musty. Sample Nineteen was a sound blend of river bottom and clay hill wheat.

The four samples showed striking variations in their amino-acid contents during tempering, in spite of the fact that they originated within a twenty-five mile radius. Sample Fourteen had practically no change in amino-acids during the entire ninety-two hours. Sample Fifteen was constant for the first twenty hours, and then decreased for the remainder of the period. Sample Seventeen, the badly sprouted wheat, decreased steadily in amino-acid content during the entire time. Sample Nineteen showed an initial increase, then a decrease for the rest of the period.

The water soluble nitrogen content of Sample Fourteen decreased during the first twenty hours, in contrast

to an increase in Samples Seventeen and Nineteen. During the latter portion of the tempering period, the water soluble nitrogen decreased at approximately the same rate in all samples except Number Seventeen, the damaged one. Sample Seventeen showed no change at ninety-two hours over the water soluble nitrogen content at twenty hours.

The water soluble nitrogen showed a positive correlation with the titratable acidity in all cases.

All the samples except Number Seventeen showed a decrease in copper non-precipitable nitrogen during the first twenty hours of the tempering period. Sample Seventeen had an increase during the early stages which was continued throughout the entire ninety-two hours. Sample Nineteen also had a similar increase during the last seventy-two hours, while samples Fourteen and Fifteen did not change appreciably after twenty hours.

These results show that the prairie and clay hill wheat are very stable during tempering, with no tendency for proteins or even poly-peptides to break down. The opposite reaction of building up higher molecular compounds is present, but is not pronounced. These two samples were the most stable of any studied. The bottom land wheat was very irregular and showed both reactions at work in the same samples. These rather inconsistent results are probably caused by the unusual climatic conditions during the harvest of 1928, when there were alternately heavy rains and hot sunshine upon the shocked wheat, delaying threshing from two to six weeks beyond normal.

TABLE 4

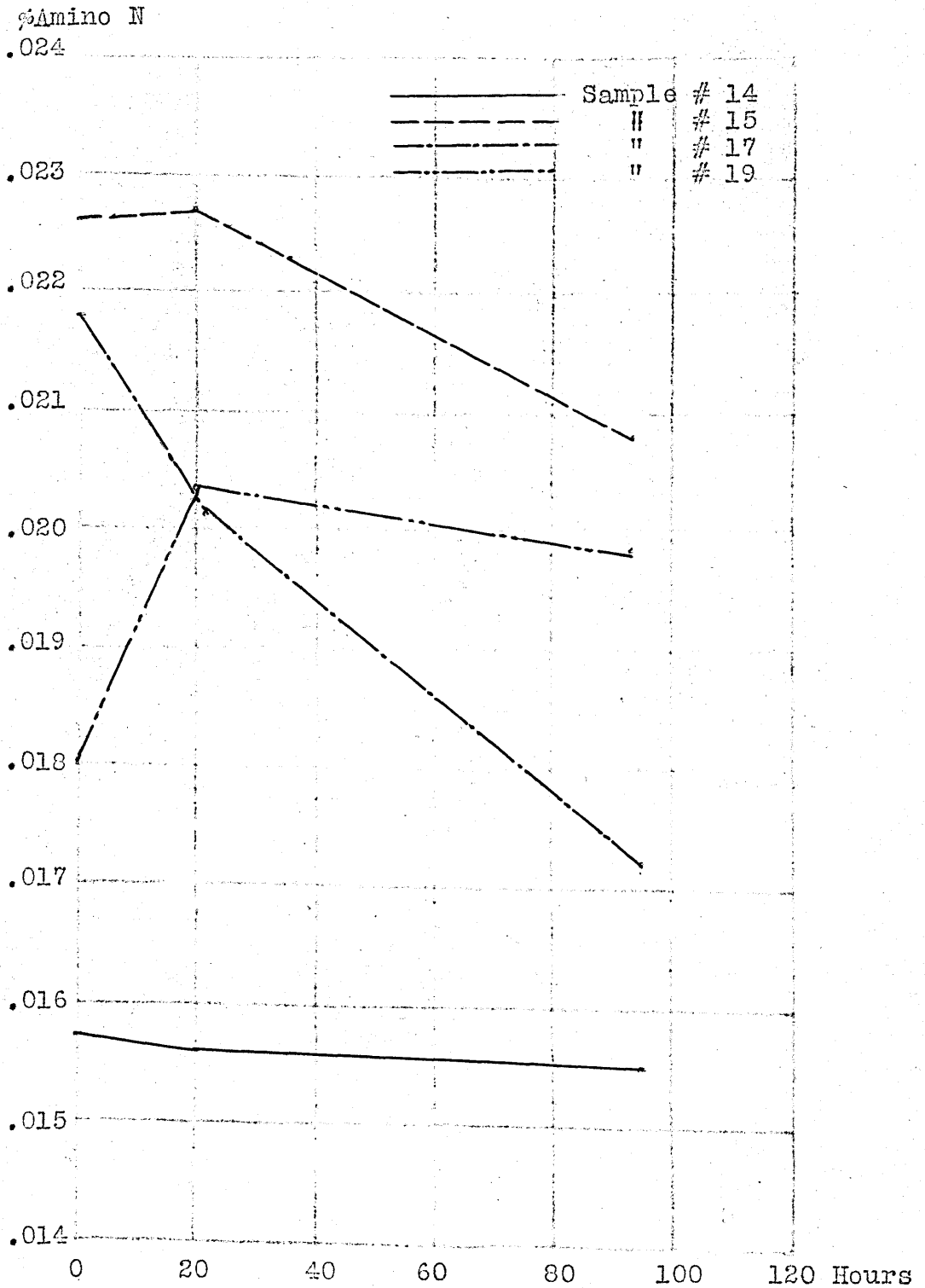
Soft Red Winter Wheat

Sample Number	Time Temp.	Moisture %	Acidity c.c. H ₂ SO ₄	Amino-N %	Water Sol.N %	Cu Non- pp.N %
14	0	9.14	2.85	.0157	.330	.0788
14	20	16.22	2.6	.0156	.320	.0740
14	92	15.40	2.15	.0155	.303	.0734
15	0	10.88	3.35	.0226	.371	.0964
15	20	16.28	3.45	.0227	.372	.0910
15	92	16.82	3.3	.0208	.354	.0916
17	0	15.24	2.95	.0218	.338	.0928
17	21	17.32	3.6	.0201	.347	.0980
17	92	17.89	4.1	.0172	.347	.1044
19	0	10.63	3.4	.0180	.321	.0889
19	20	18.18	3.85	.0204	.346	.0846
19	92	18.88	3.8	.0198	.325	.1000

All results except moisture are calculated to 15% moisture basis.

Soft Red Winter Wheat

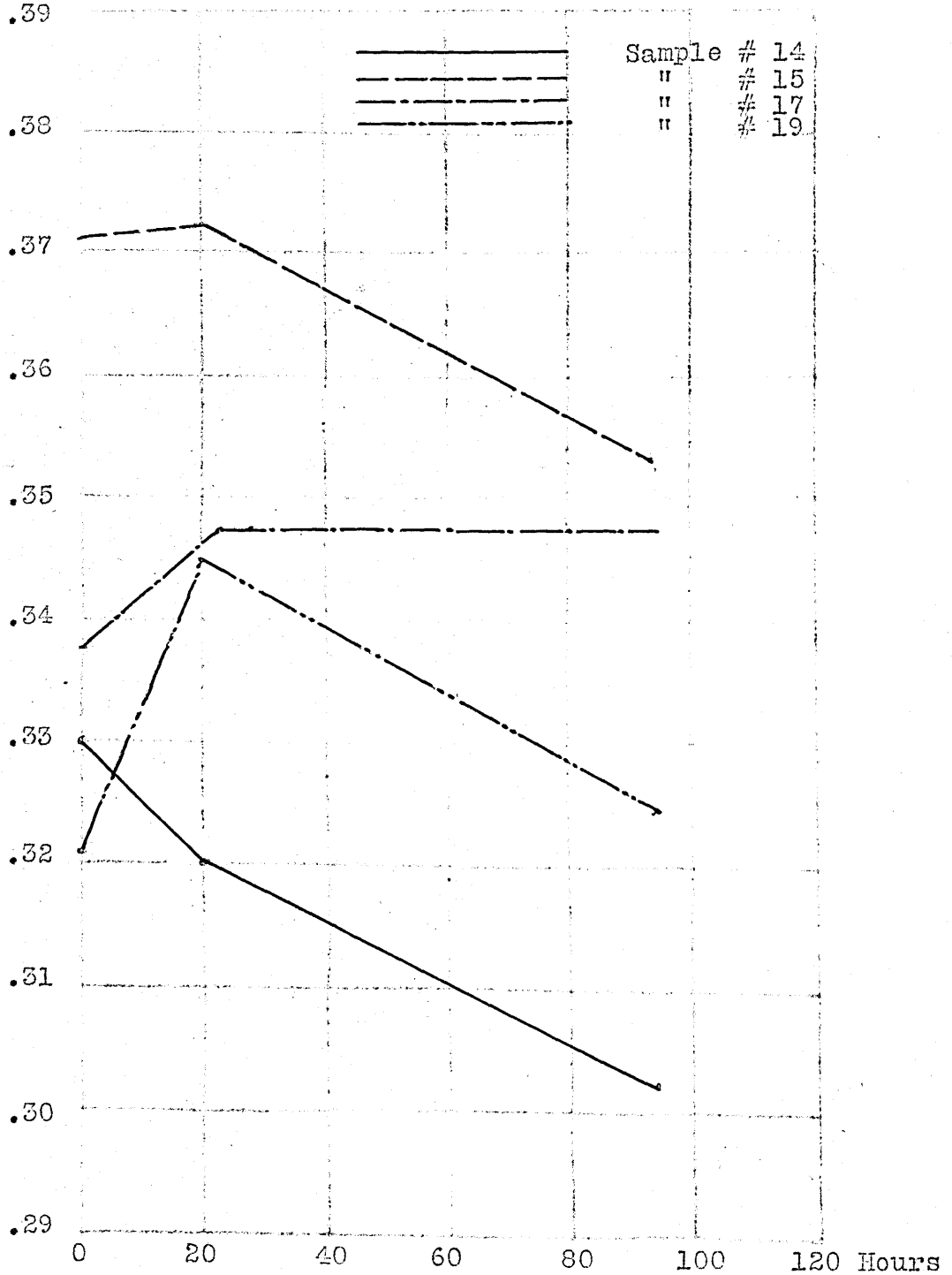
Amino Nitrogen - Tempering Time.



Soft Red Winter Wheat

Water Soluble Nitrogen - Tempering Time

% Water Sol. N.

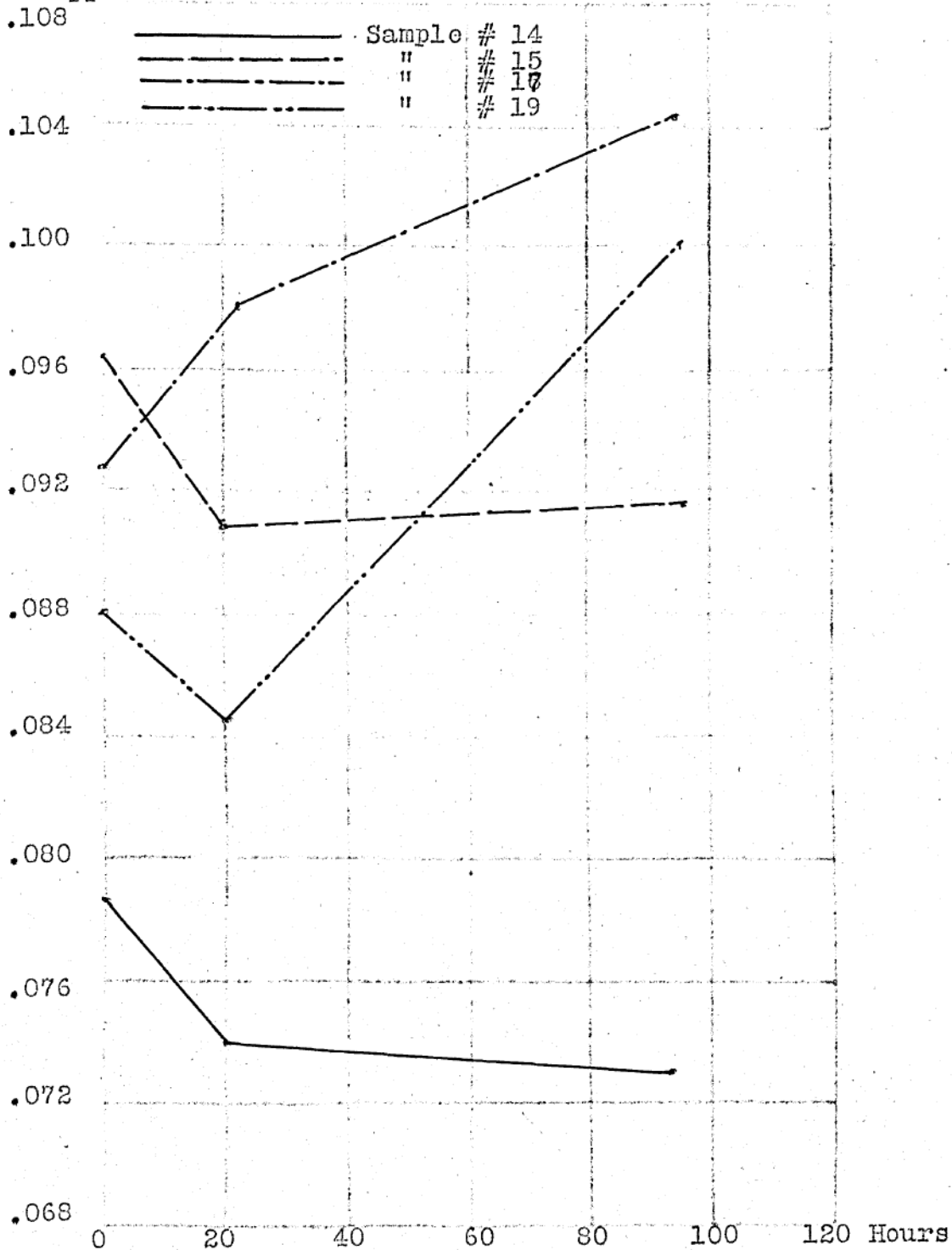


Soft Red Winter Wheat

Copper Non-precipitable Nitrogen -

Tempering Time

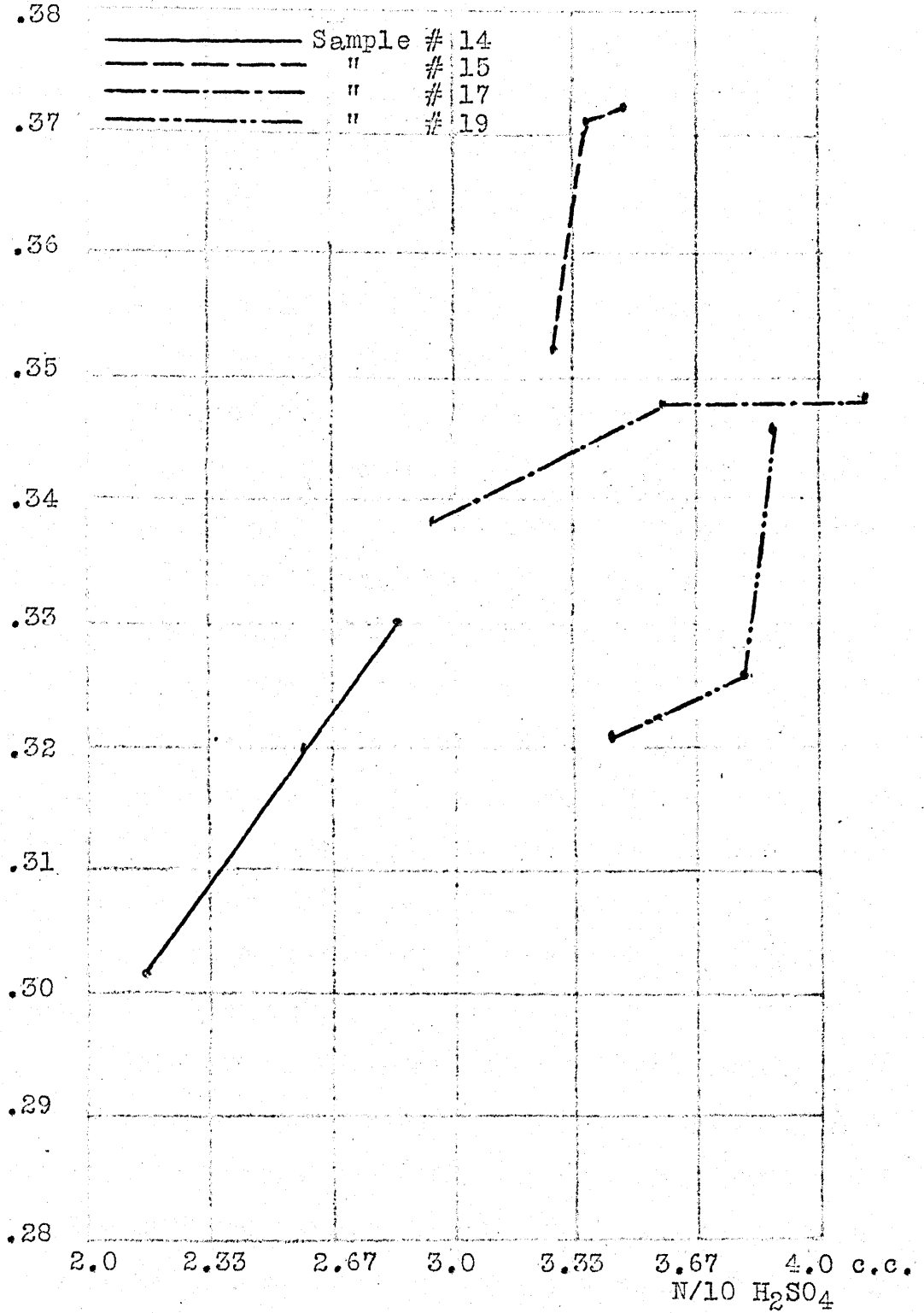
%Cu-non-pp N.



Soft Red Winter Wheat.

Water Soluble Nitrogen - Titratable Acidity..

Water Sol.N.



Four samples of hard red winter wheat were chosen for this work. They were country-run samples from southwestern Kansas. They all originated in a strip of country about six miles wide, extending about thirty miles south from Garfield in Pawnee County, on the Arkansas River. The total nitrogen content of the samples was very uniform, only varying eight-hundredths of one per-cent between the extremes.

Three of the four samples showed a decided increase in amino-acid content during the first twenty hours, and a decrease during the latter part of the time. This indicated that during the first twenty hours the moisture content was sufficient to start the germination process, but not enough to sustain it. The fourth sample, Number Sixteen, varied from the others in that the amino-acid content fell off sharply during the first twenty hours, and then did not change. This showed that the moisture added was insufficient to even start germination in this sample, and that desiccation set in at once, but soon reached a limiting value.

The water soluble nitrogen content closely paralleled the amino-acid content, with the exception that in sample Sixteen the water soluble nitrogen continued to fall off at the same rate during the entire time. As this rate closely paralleled the rate of decrease during desiccation

for the other three samples, it may be considered as additional proof that desiccation set in from the start with this sample.

The copper non-precipitable nitrogen content during the first twenty hours behaved similarly to both the amino-acid and the water soluble nitrogen contents. But after the first twenty hours, samples Eleven and Eighteen decreased sharply, while samples Ten and Sixteen increased at about the same rate as the others decreased. This indicates that a secondary reaction had set in, possibly a splitting of the higher peptide linkings, forming simpler poly-peptides. The only reason that can be found for this difference is that samples Ten and Eighteen were grown upon sandy soil, while the other two were from hard land.

The samples other than Sixteen show a positive though erratic correlation between the water soluble nitrogen and the titratable acidity.

TABLE 5

Hard Red Winter Wheats.

Sample Number	Time Temp. Hrs.	Moisture %	Acidity c.c. H ₂ SO ₄	Amino-N %	Water Sol. N %	Cu Non- pp. N %
10	0	10.78	2.45	.0187	.307	.0696
10	20	16.06	3.05	.0213	.341	.0856
10	92	16.54	2.75	.0164	.333	.0912
11	0	9.59	2.95	.0197	.327	.0668
11	20	16.44	3.25	.0214	.352	.0856
11	92	15.60	2.8	.0191	.333	.0708
16	0	10.28	2.05	.0189	.318	.0850
16	20	15.97	2.95	.0138	.307	.0768
16	92	18.40	2.4	.0138	.272	.0848
18	0	9.06	3.35	.0177	.316	.0840
18	22	16.86	3.05	.0193	.337	.0890
18	94	16.37	2.6	.0171	.304	.0828

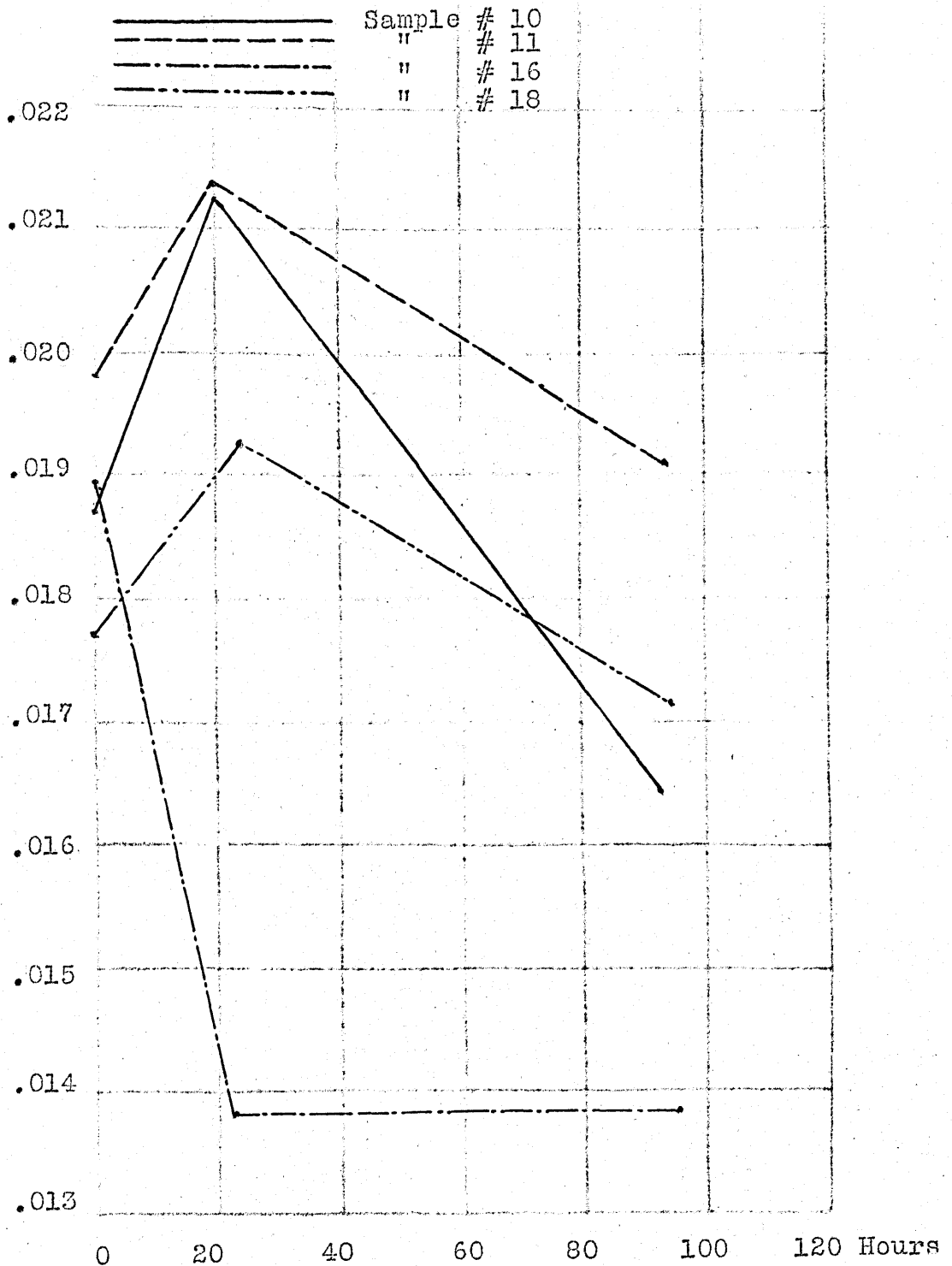
All results except moisture are calculated to 15% moisture basis.

Hard Red Winter Wheat

Amino-Nitrogen - Tempering Time

%Amino-N.

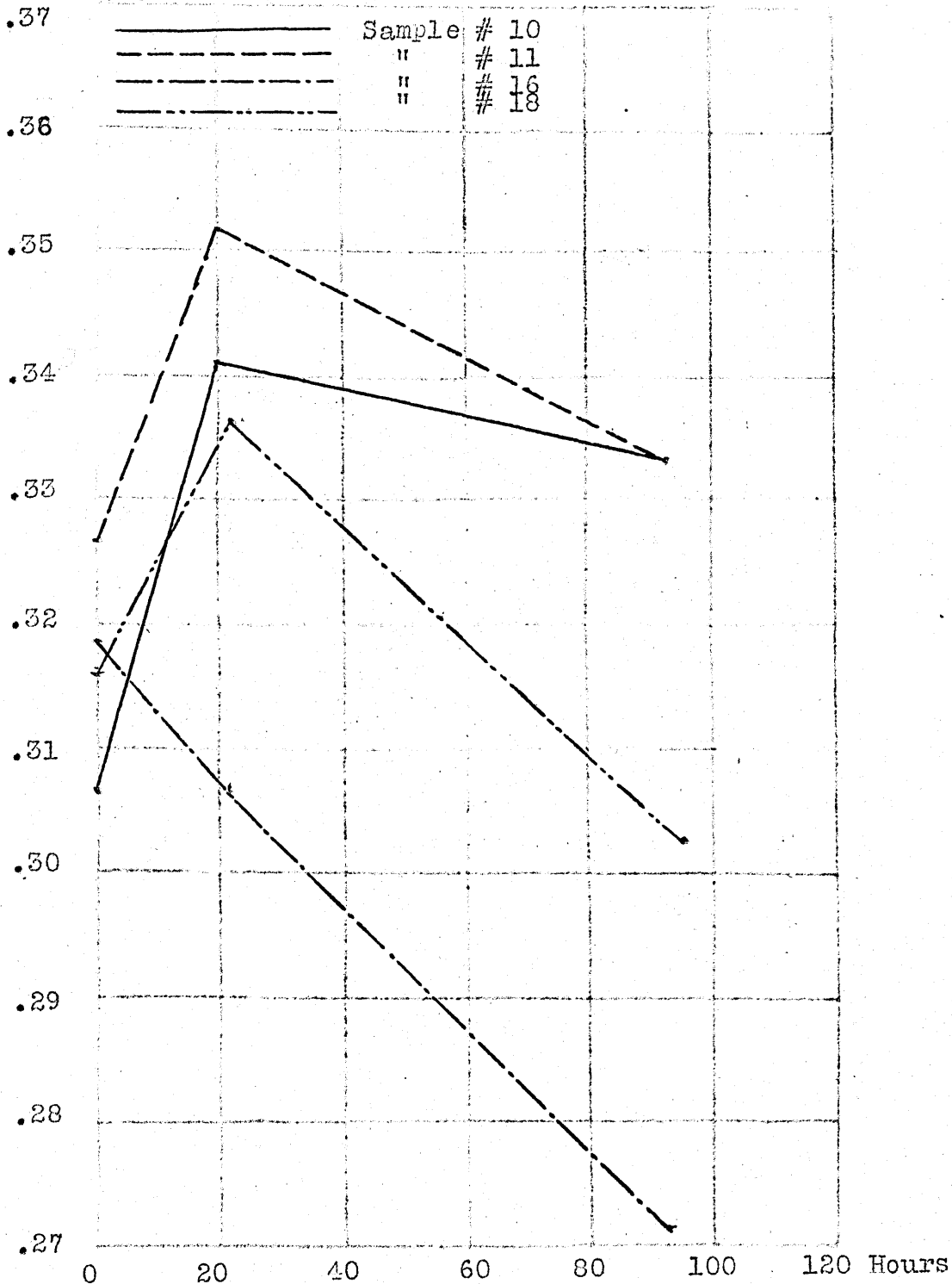
.023



Hard Red Winter Wheats

Water Soluble Nitrogen - Tempering Time

% Water Sol.N.

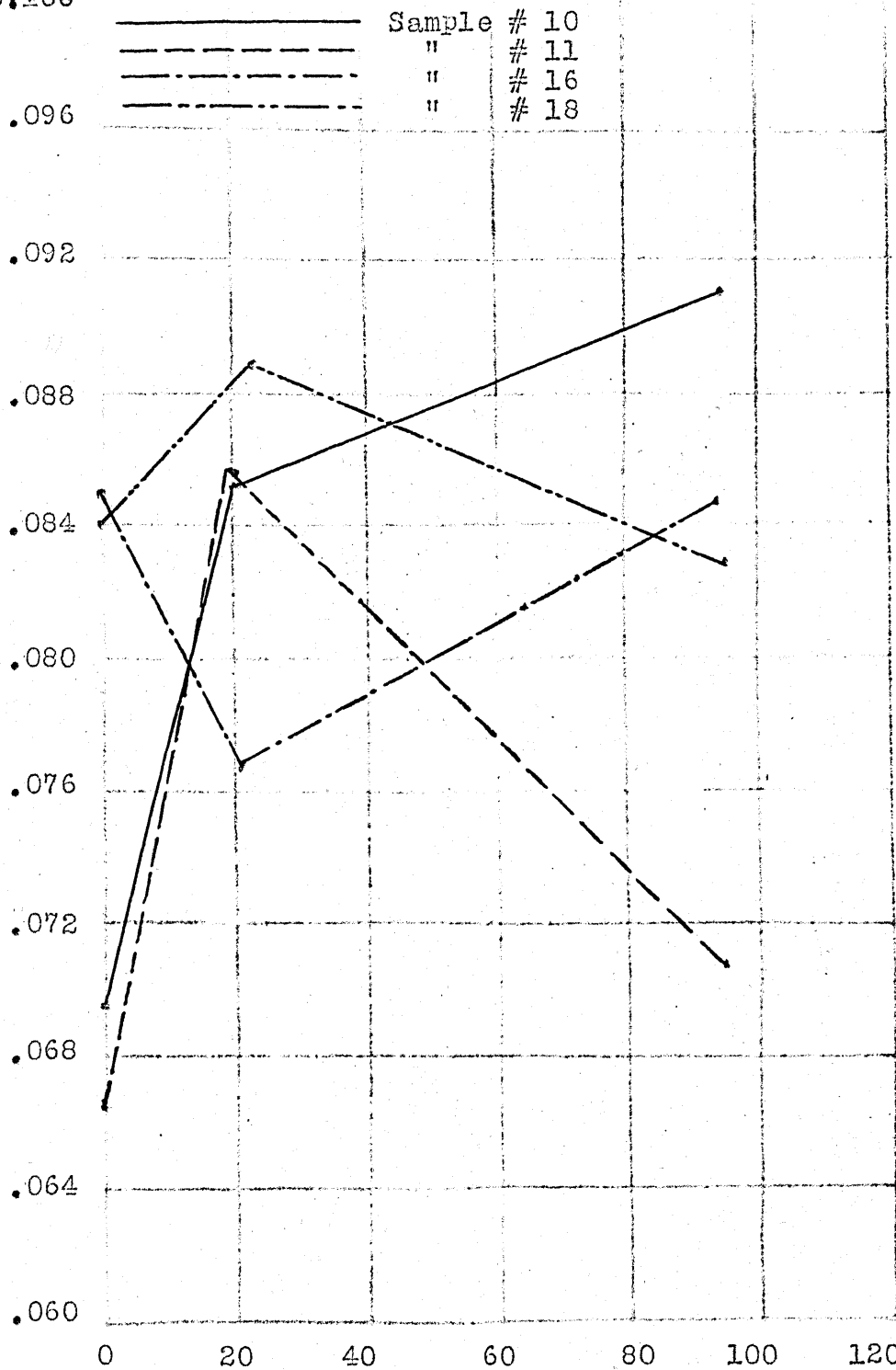


Copper Non-precipitable Nitrogen - Tempering Time

Hard Red Winter Wheats

%Cu non-p.p.N.

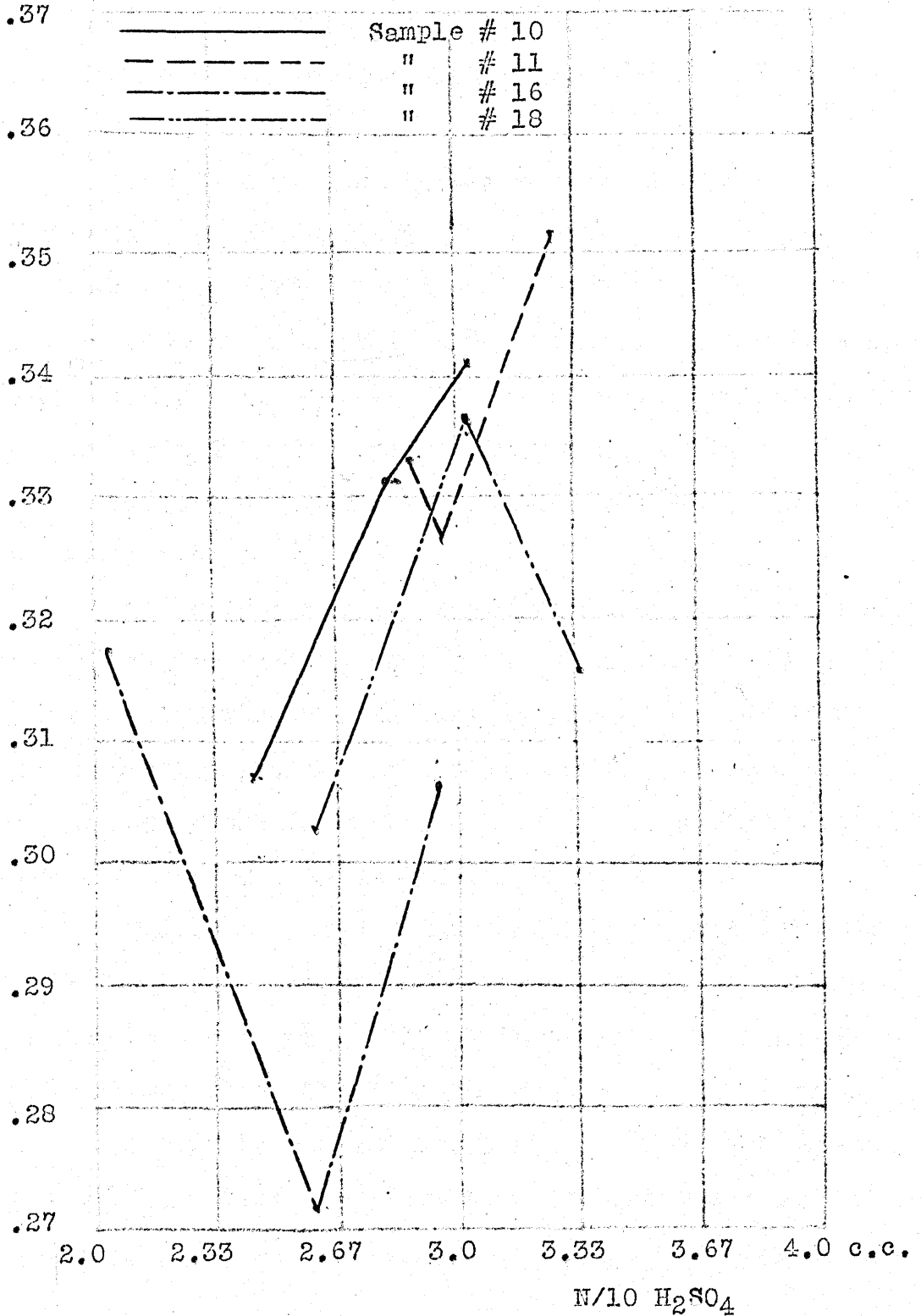
0.100



Hard Red Winter Wheat

Water Soluble Nitrogen - Titratable Acidity

%Water Sol.N.



Two samples of Dicklow wheat were studied. One was a sample of an Ogden, Utah, elevator blend, and the other was a car sample of country-run wheat from Buhl, Idaho.

The Utah sample, Number Twelve, gave a decided increase in amino-acids during the first twenty hours. After that, desiccation set in and there was a decrease in amino-acids during the rest of the tempering period. The Idaho sample showed a lesser increase in amino-acids during the first twenty hours, but during the remainder of the tempering period there was a continued increase, though at a slower rate.

The copper non-precipitable nitrogen in the Utah sample increased rapidly during the first twenty hours, and then fell off nearly as rapidly during the remainder of the tempering period. The Idaho sample showed a slight increase during the first twenty hours, but a rapid increase during the last portion of the tempering period.

While the water soluble nitrogen was but little affected by the tempering, the curves in general followed the amino-acid changes. The correlation between the water soluble nitrogen and the titratable acidity was slight.

The Idaho sample showed that in the early stages of tempering, the lower molecular poly-peptides were broken up and free amino-acids were readily formed. During the

latter part of the period this reaction slows down, and there is a tendency for higher poly-peptides and even proteins to be broken down into the lower poly-peptide forms. The moisture content of this sample after tempering was enough to delay the setting in of desiccation.

The Utah sample showed first the peptide splitting, and then the setting in of desiccation, with a reversal of the reaction.

TABLE 6

53

Dicklow Wheat

Sample Number	Time Temp. Hrs.	Moisture %	Acidity c.c. H ₂ SO ₄	Amino-N %	Water Sol. N %	Cu Non- pp. N %
12	0	10.69	2.33	.0200	.308	.0854
12	20	17.33	2.62	.0231	.354	.0922
12	86	17.35	2.67	.0216	.343	.0838
13	0	10.36	2.42	.0213	.308	.0744
13	20	17.56	2.17	.0224	.330	.0752
13	86	16.80	2.76	.0229	.333	.0888

Dicklow Wheat.

Amino-Nitrogen - Tempering Time

%Amino-N.

.027

.026

.025

.024

.023

.022

.021

.020

.019

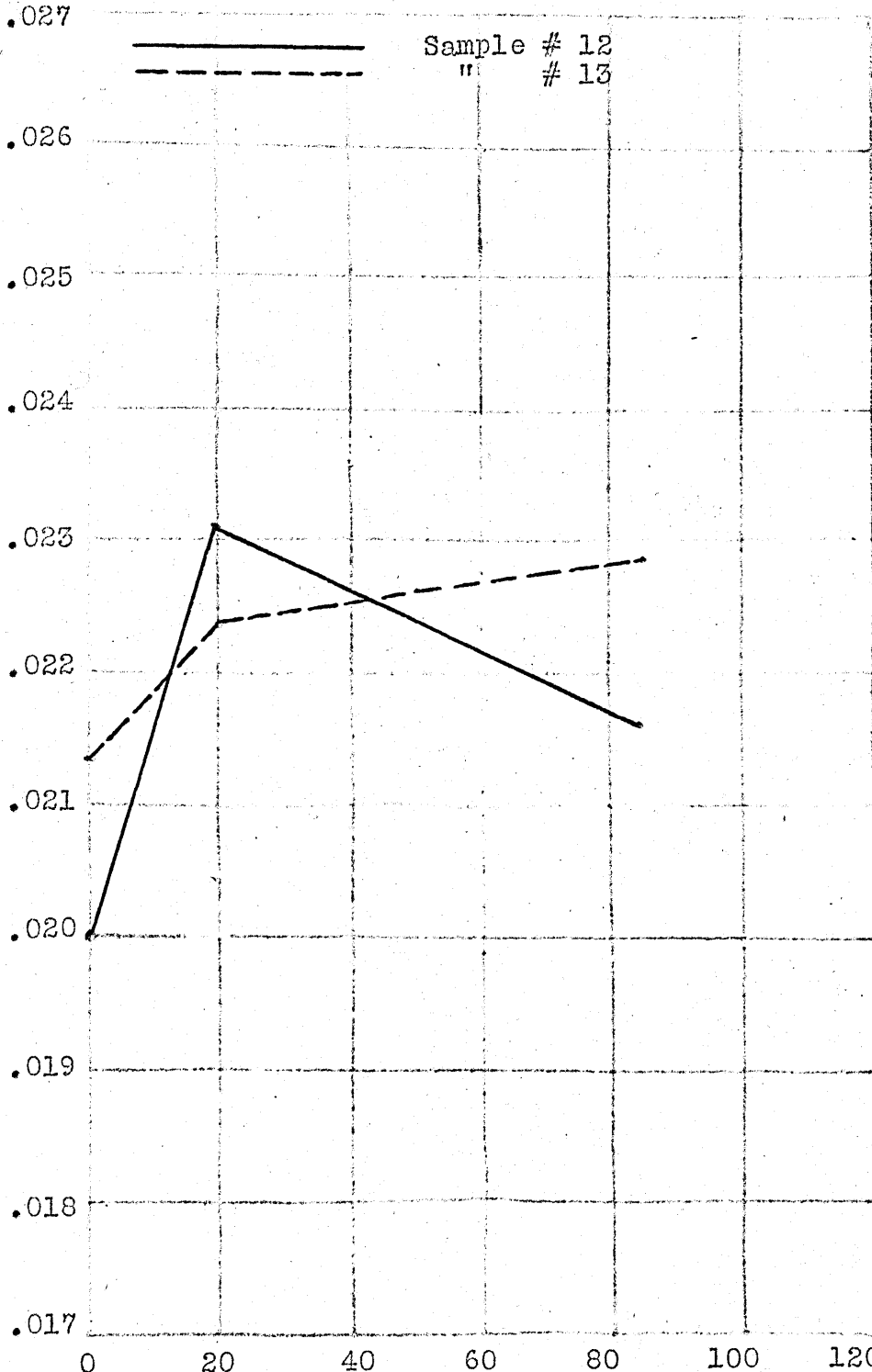
.018

.017

Sample # 12

" # 13

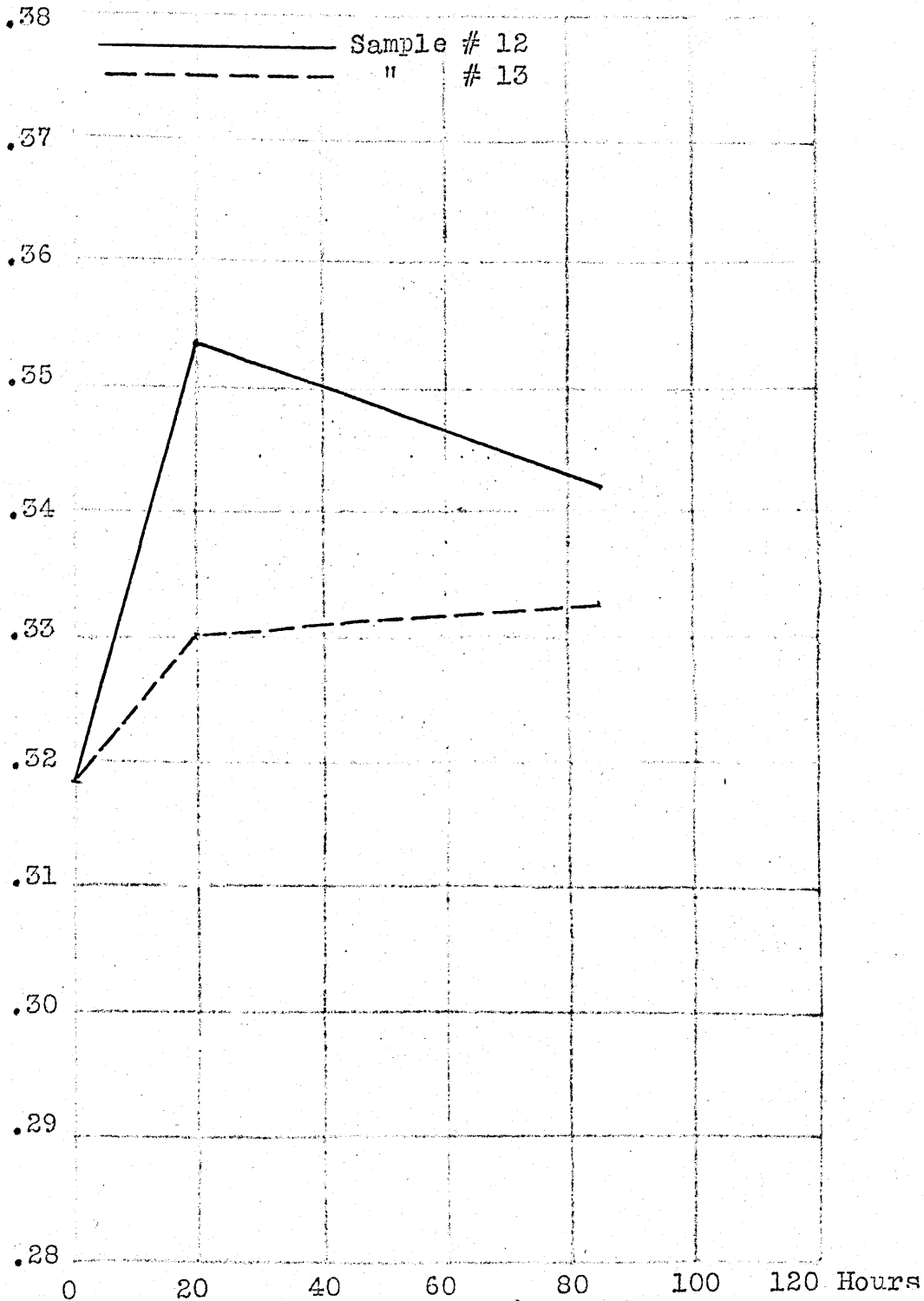
0 20 40 60 80 100 120 Hours



Dicklow Wheat.

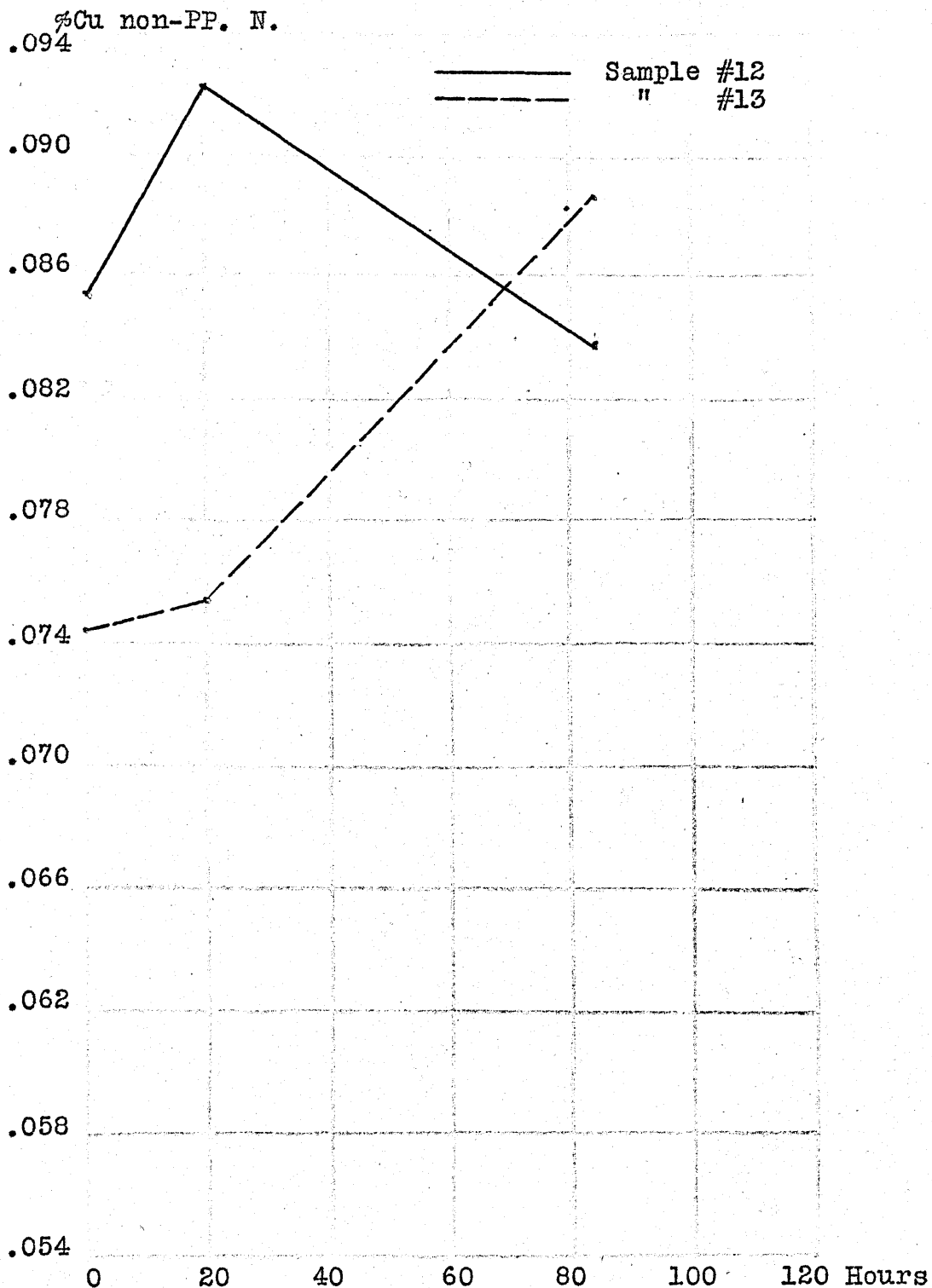
Water Soluble Nitrogen - Tempering Time.

%Water Sol.N.



Dicklow Wheat

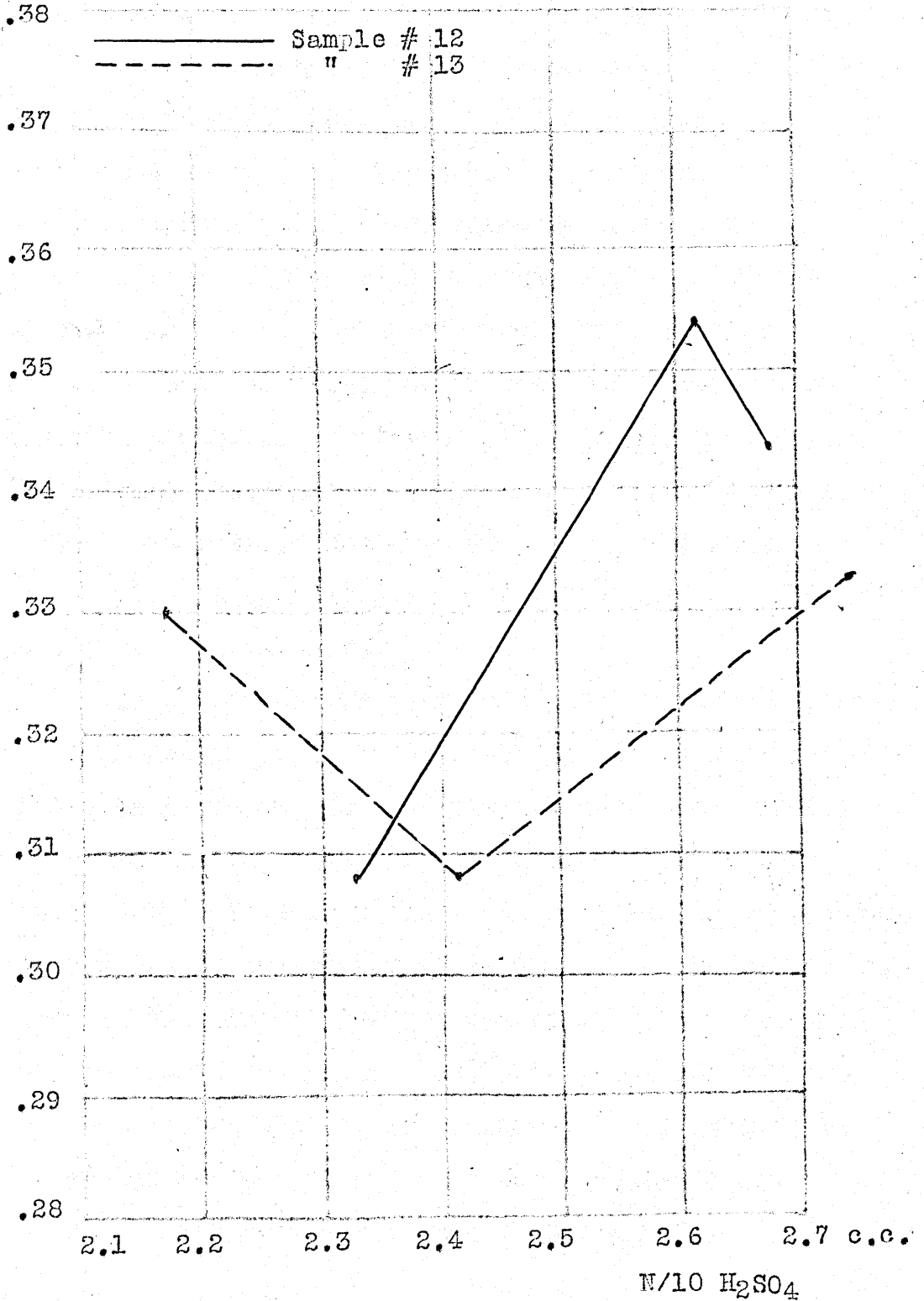
Copper Non-precipitable Nitrogen - Tempering Water.



Dicklow Wheat

Water Soluble Nitrogen - Titratable Acidity.

% Water Sol. N.



Three samples of Soft Federation Wheat were used in this work. Sample Number Three came from Fulton station in the Willamette Valley, about four miles south of Portland, Oregon. This is a humid region with no extremes in temperature. The other two samples were from Umatilla County in the northeastern corner of the state of Oregon. Number Four was from Helix, and Number Five from Vansycle, two stations about fifteen miles apart. This part of Oregon is in the great inter-mountain basin, a section of plains taking in eastern Oregon, southern Idaho, and much of Utah, and having cold winters and hot, dry summers. The greater part of the wheat raised in this section is grown under irrigation, although a small part is dry-farmed.

Sample Number Three from the Willamette Valley showed a slight increase in amino-acid content during the first twenty hours of the tempering period. But after twenty hours there was no further change in amino-acid content. Sample Four from Helix, in Umatilla County, showed a larger increase during the first twenty-one hours. During the rest of the time it had a sharp decrease to less than the original value at ninety-two hours. The sample from the neighboring station of Helix showed little change in the first fifteen hours, and then the amino-acid content increased sharply during the rest of the period.

The water soluble protein content of sample Number Three increased sharply during the first twenty hours, then fell off at nearly as rapid a rate during the last seventy-two hours. Both samples from Umatilla County showed slight increases during the first period. The sample from Helix station continued to gain the entire time, while the Vansycle sample reduced at about the same rate as the Willamette Valley sample.

The Soft Federation Wheat samples showed a slight correlation between the water soluble nitrogen and the titratable acidity.

Sample Number Three from Fulton in the Willamette Valley had an initial decrease during the first twenty hours in copper non-precipitable nitrogen, and later a large increase. The sample from Helix in Umatilla County had a moderate initial increase during the first period, with continued increase at a slower rate during the latter period. The Vansycle sample showed a very large initial increase in copper non-precipitable nitrogen, followed by a sharp decrease after twenty hours.

The sample from the humid Willamette Valley was a little more stable than the samples from the irrigated section. The two samples from Umatilla County had decidedly different reactions during the tempering. These differences were probably due to cultural variances, or

to the presence of dry farming wheat in one of the samples. There was so large a variation in the different Federation samples that no distinctive varietal factors in the changes in nitrogen distribution during tempering can be noted.

TABLE 7

61

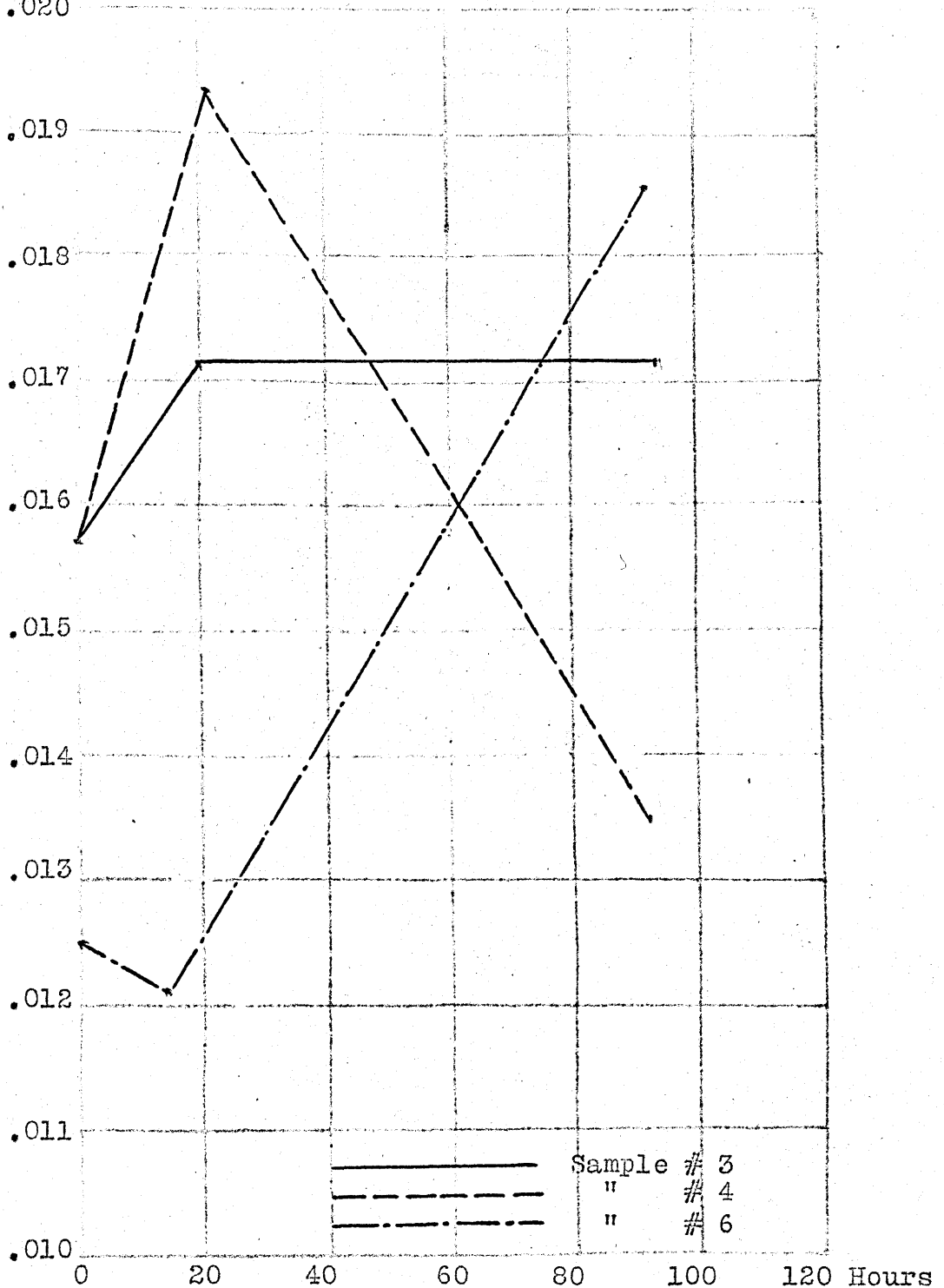
Soft Federation Wheat

Sample Number	Time Temp. Hrs.	Moisture %	Acidity c.c. H ₂ SO ₄	Amino-N %	Water Sol. N %	Cu Non- pp. N %
3	0	8.94	2.4	.0157	.272	.0708
3	20	16.98	2.85	.0172	.312	.0690
3	92	16.99	2.25	.0172	.282	.0776
4	0	9.30	2.4	.0157	.290	.0658
4	21	17.13	2.25	.0194	.306	.0722
4	92	16.15	2.55	.0135	.314	.0740
6	0	9.71	3.15	.0125	.327	.0582
6	15	16.44	3.4	.0121	.339	.0772
6	92	16.69	2.45	.0186	.316	.0716

Soft Federation Wheat

Amino-Nitrogen - Tempering Time

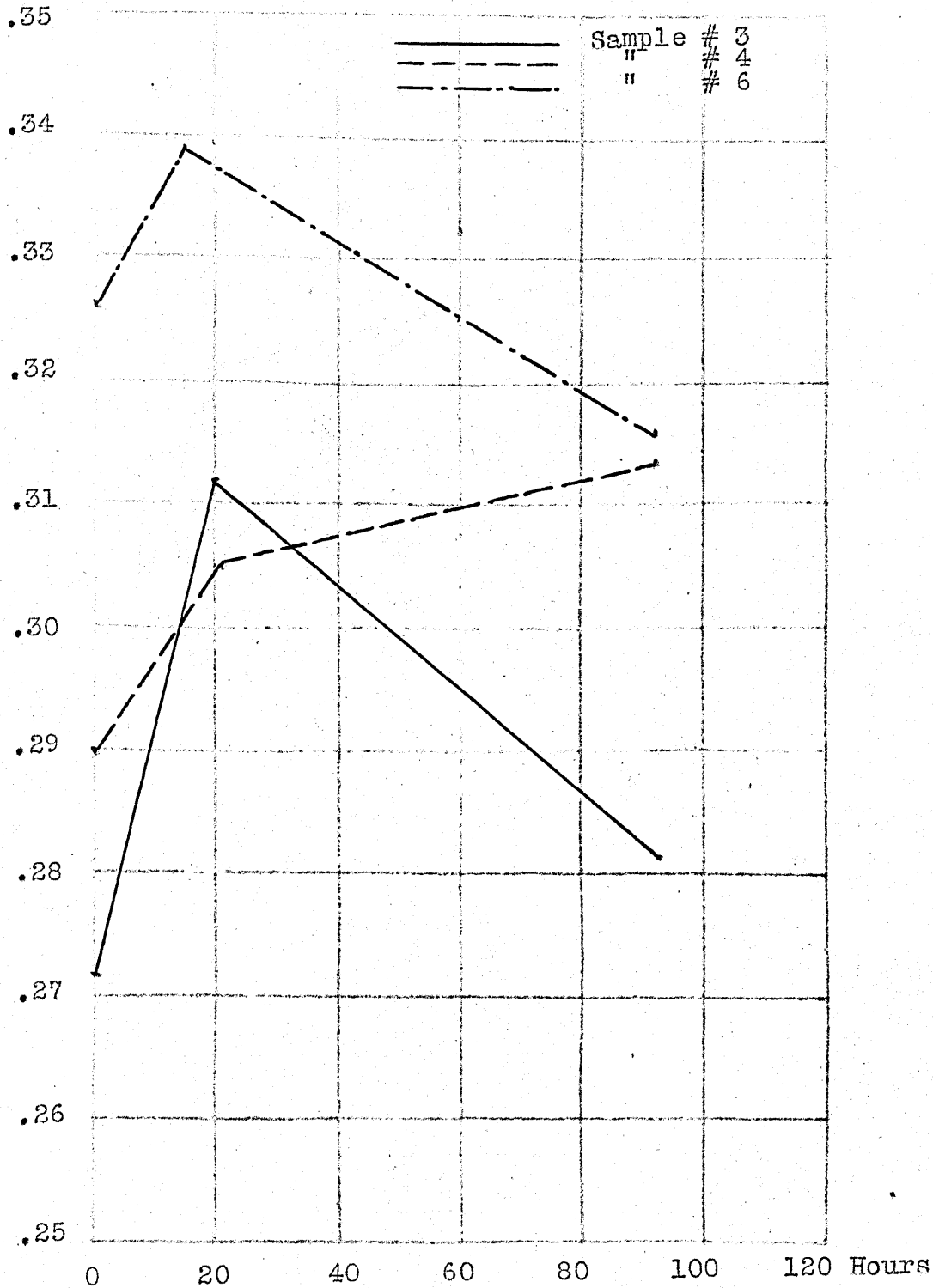
% Amino-N.
.020



Soft Federation Wheat

Water Soluble Nitrogen - Tempering Time

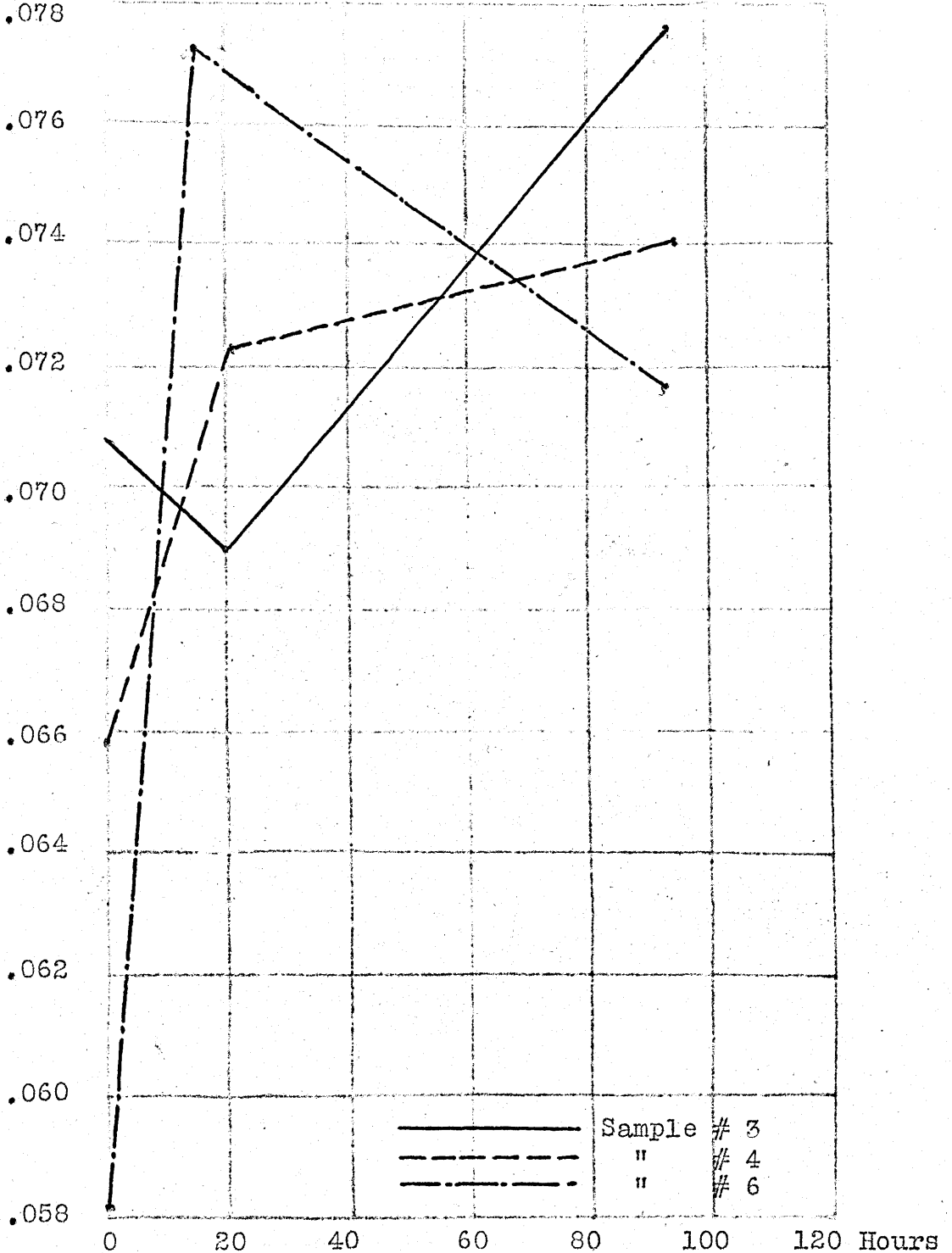
% Water Sol. N.



Soft Federation Wheat

Copper Non-precipitable Nitrogen - Tempering Time.

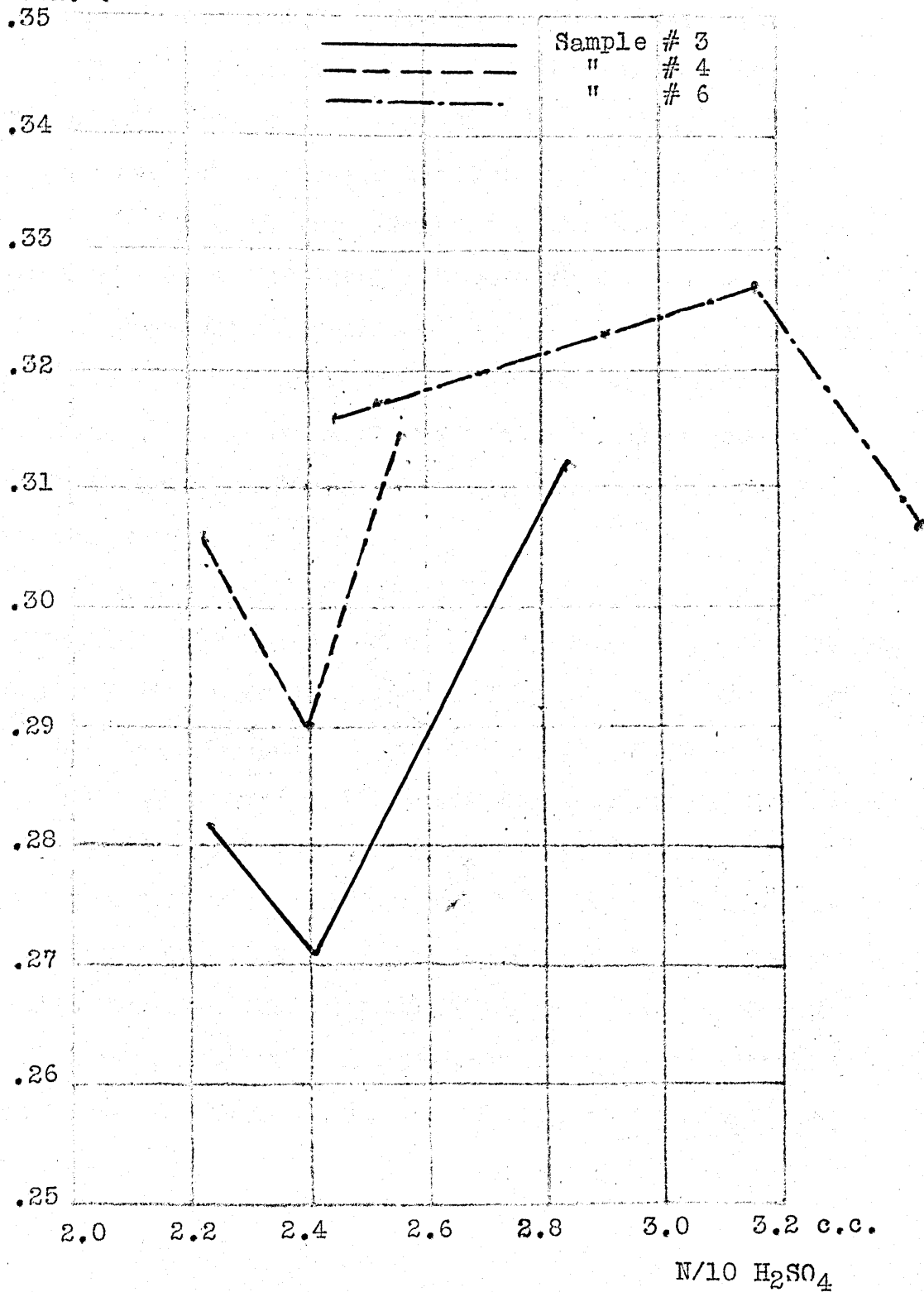
%Cu non-pp.N.



Soft Federation Wheat

Water Soluble Nitrogen - Titratable Acidity

% Water Sol.N.



Only two samples of this variety of soft white club wheat were available. One sample was from Fulton station in the Willamette Valley of Oregon, and the other was from Vansycle in Umatilla County, Oregon. This made both samples directly comparable with samples of Soft Federation wheat from the same stations. These samples ran much lower in total nitrogen than the Soft Federation samples from the same stations. These two samples had the lowest total nitrogen content of any of the samples studied, being about three-quarters of one per-cent lower than the highest sample, which was soft red wheat, Number Fifteen.

Both samples of Hybrid 128 wheat showed a moderate initial increase in amino-acids. After the first twenty hours, the Fulton sample dropped off to a final value slightly lower than the original. In contrast, the sample from Umatilla County showed a continued increase during the final period. The general appearance of the amino-acid curves for the two Fulton samples was similar. This was likewise true of the Vansycle samples. The samples from Fulton in the Willamette Valley were relatively stable and showed little change, while the Umatilla County samples showed a decided breaking down at the end of ninety-two hours' tempering.

The water soluble nitrogen curves for the two Hybrid 128 wheat samples ran practically parallel. There was an initial increase during the first twenty hours, then a further increase at a much slower rate. This was in contrast to the corresponding samples of Soft Federation wheat, which showed an initial increase, then a decrease. As the water soluble nitrogen is a function of the colloidal condition of the wheat, rather than a function of the chemical constitution of the sample, these slight variations may be varietal rather than environmental. This supposition is open to the criticism that the sample of Soft Federation from Helix, a neighboring station to Vansycle, showed the same water soluble nitrogen curve as the Hybrid 128 samples.

The sample of Hybrid 128 wheat from Fulton station showed a slight initial decrease in copper non-precipitable nitrogen during the first twenty hours, then an increase for the last seventy-two hours. These changes were so slight as to be within the experimental error, but as they paralleled the larger changes in the sample of Soft Federation wheat from the same station, they may have significance.

The sample of Hybrid 128 wheat from Vansycle showed a very large initial increase in copper non-precipitable nitrogen of the same relative magnitude as the Soft Federation sample from the same station. During the latter part of the tempering period there was a very slight increase,

but on the whole this sample agreed well with the Soft Federation sample from the same station. The findings of the copper non-precipitable nitrogen confirmed the amino-acid content, in that the Willamette Valley wheat was much more stable than the wheat from Umatilla County. It also confirmed the finding that the differences between these two botanically-different classes of wheat, insofar as chemical changes in tempering were concerned, were environmental, rather than varietal and inherent.

As the Dicklow variety showed similar dissimilarity in the changes in its copper non-precipitable nitrogen content, and also as the soft red winter wheats and the hard red winter wheats were not uniform with respect to variety, it may be assumed that the chemical changes in the nitrogen distribution, due to the tempering of wheat, are functions of the environment of the wheat, and are not inherent varietal functions. This assumption must not be held to include physical or colloidal changes during tempering, unless substantiated by further research work.

TABLE 8

69

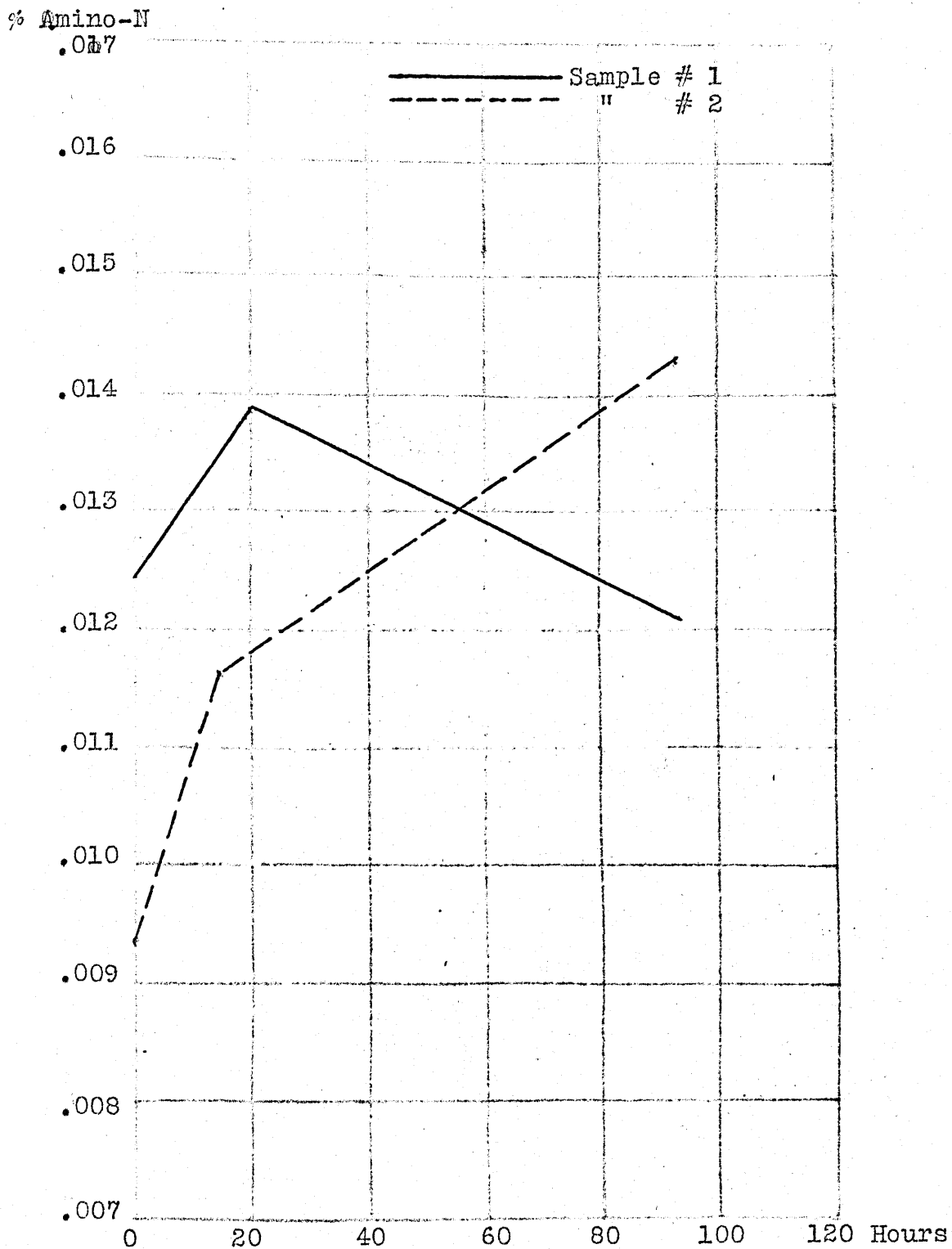
Hybrid 128 Wheat

Sample Number	Time Temp. Hrs.	Moisture %	Acidity c.c. H ₂ SO ₄	Amino-N %	Water Sol.N %	Cu Non- pp.N %
1	0	9.10	2.4	.0124	.254	.0598
1	20	17.37	2.3	.0138	.267	.0578
1	92	16.67	2.4	.0121	.276	.0602
2	0	10.47	3.0	.0094	.265	.0434
2	15	17.54	3.9	.0116	.284	.0724
2	92	16.80	2.85	.0143	.290	.0746

All results except moisture are calculated to 15% moisture basis.

Hybrid 128

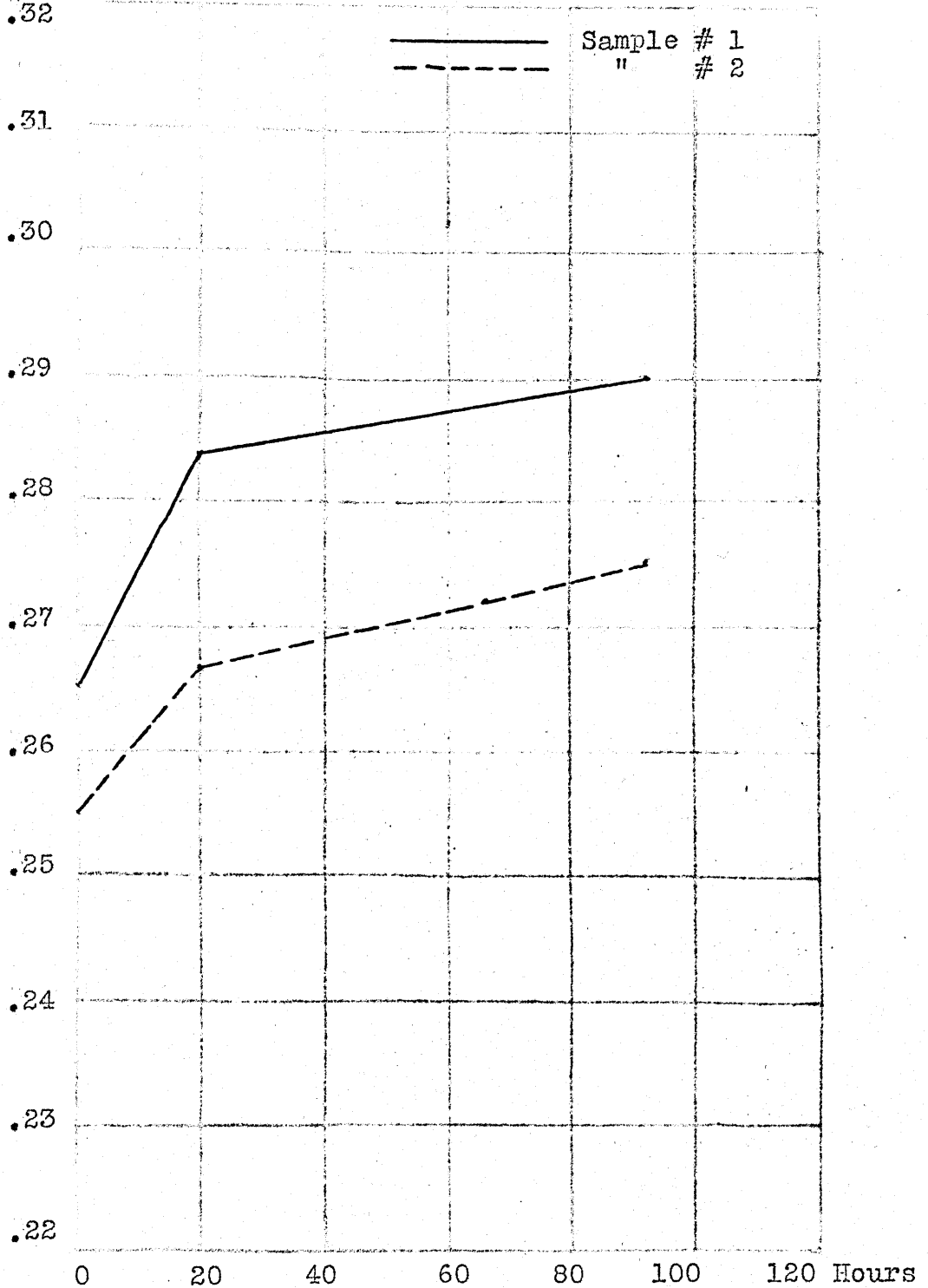
Amino-Nitrogen - Tempering Time



Hybrid 128 Wheat

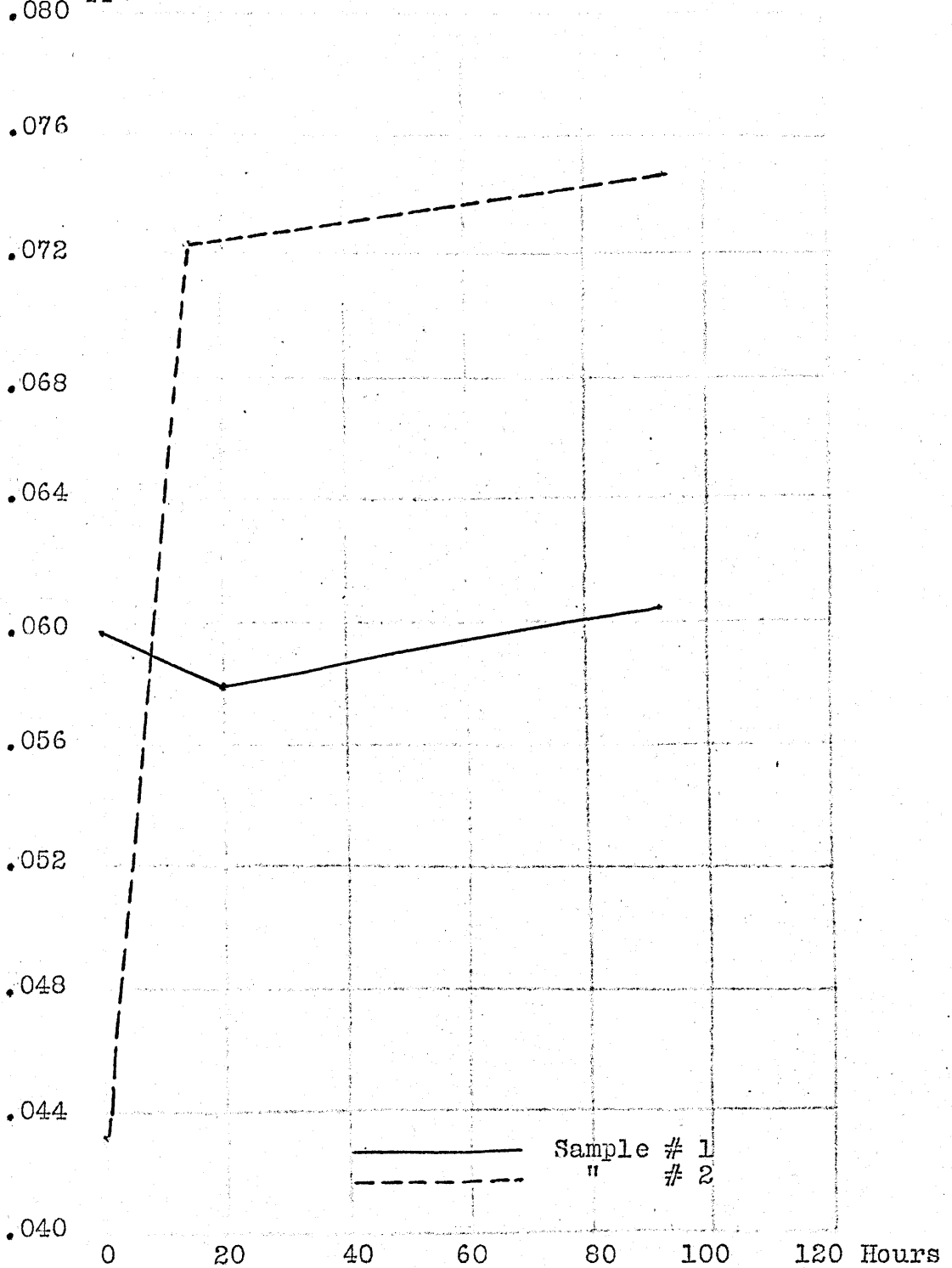
Water Soluble Nitrogen - Tempering Time.

% Water Sol. N.



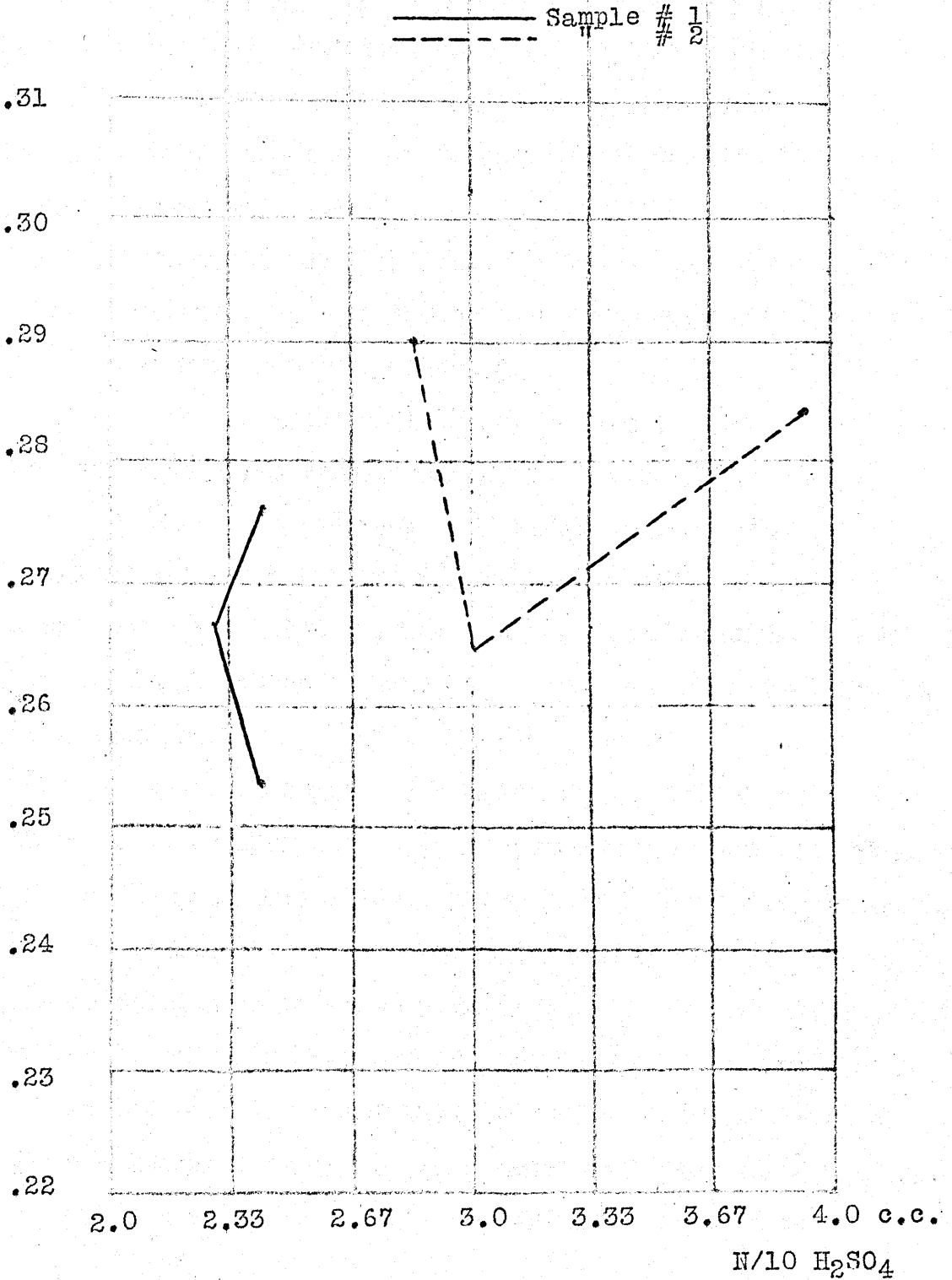
Hybrid 128 Wheat

Copper Non-precipitable Nitrogen - Tempering Time.

% Cu non-pp.N.
.080

Hybrid 128 Wheat.

Water Soluble Nitrogen - Titratable Acidity.

% Water Sol.N.
.32

VARIABLE HYDROGEN ION CONCENTRATION
OF THE TEMPERING WATER.

74

One sample of each type of wheat was selected for this series. Sample Number One, Hybrid 128, came from the Willamette Valley in Oregon. Sample Number Four, Soft Federation, was from Umatilla County, Oregon. Sample Number Ten, low protein hard winter wheat, was grown at Garfield, in the Arkansas River bottom of southwestern Kansas. Sample Number Nineteen, soft red winter wheat, was from central Missouri.

As the variation in the moisture content of the dry wheats was small, portions of each sample were tempered with ten per-cent of the following solutions: distilled water, tenth normal sulfuric acid, hundredth normal sulfuric acid, tenth normal sodium hydroxide, hundredth normal sodium hydroxide, and twentieth normal sodium sulfate solution.

The first point of interest was to determine the effect of the different tempering solutions upon the titratable acidity of the wheats, as compared to titratable acidity of the samples tempered with distilled water. The twenty hour results were very consistent. In all four samples the tenth normal sulfuric acid increased the titratable acidity to about the calculated amount. The tenth normal sodium hydroxide, however, only decreased the acidity of the soft red winter wheat about seventy-five per-cent of the

calculated amount, and the hard red winter wheat about forty per-cent. On the Oregon white wheat samples the tenth normal sodium hydroxide gave the same results as the distilled water, indicating that all of the alkali had gone into chemical composition with the wheat.

The hundredth normal and the salt solutions were rather erratic. At ninety-two hours the results were much the same as at twenty hours, with no consistent changes. The sodium hydroxide probably combined with the proteins, but some may have saponified a portion of the fatty bodies in the embryo. The samples tempered with the tenth normal sodium hydroxide were much slower filtering than the other samples.

The Hybrid 128 wheat showed an increase in the amino-acid content during twenty hours' tempering with distilled water. The sample was too small to permit tempering with all solutions to ninety-two hours. The hundredth normal sodium hydroxide solution gave a very slightly greater increase in amino-acids, while the hundredth normal sulfuric acid and the twentieth normal sodium sulfate solution gave lesser increases. The tenth normal sulfuric acid and sodium hydroxide gave decided decreases during the same period of time.

The sample of Soft Federation wheat had a large increase in amino-acids during the first twenty hours upon water tempering, then a still larger decrease during the second portion of the tempering period. The hundredth

normal solutions gave nearly as great increases during the first period, but were constant during the last seventy-two hours. The twentieth normal sodium sulfate tempered sample showed a small decrease during the first twenty hours, but increased later to about the same figure as the hundredth normal sodium hydroxide. The tenth normal sulfuric acid and the tenth normal sodium hydroxide caused the amino-acid content to decrease exactly the same amount in twenty hours, slightly more than the sodium sulfate sample. By ninety-two hours, the samples tempered with the tenth normal sulfuric acid and the tenth normal sodium hydroxide had increased in amino-acids to about the same figure as the samples tempered with the hundredth normal solutions.

The soft red winter wheat sample, upon water tempering, showed an increase in amino-acids during the first twenty hours, then a very small decrease. When tenth normal sodium hydroxide was used instead of distilled water, there was a large increase in the amino-acids, and then a falling off during the latter period to about the same value as the water. Tenth normal sulfuric acid gave only slightly lower results than the distilled water during tempering. The twentieth normal sodium sulfate solution showed no change during the first twenty hours, then increased to about the same value as water at ninety-two

hours. The hundredth normal sulfuric acid and sodium hydroxide solutions at ninety-two hours gave about the same results as water upon the amino-acid content of this sample.

The hard winter wheat sample behaved similar to the Soft Federation sample when tempered with water, showing a large increase in amino-acids during the first twenty hours, then a still larger decrease during the last seventy-two hours. At twenty hours, all the acid, alkali, and salt solutions lowered the amino-acid content. Hundredth normal sulfuric acid showed the least decrease. Tenth normal sulfuric acid, tenth normal sodium hydroxide, twentieth normal sodium sulfate, and hundredth normal sodium hydroxide showed successive decreases in amino-acids. At ninety-two hours, the amino-acids were about evenly spaced in values, with hundredth normal sulfuric acid at the top, then in decreasing order: tenth normal sulfuric acid, hundredth normal sodium hydroxide, distilled water, twentieth normal sodium sulfate, and tenth normal sodium hydroxide at the bottom.

These results show that the behavior of amino-acids upon tempering is a function of the individual sample, rather than the hydrogen ion concentration of the tempering water. But when the results are collected together, certain general differences stand out. Samples tempered with distilled water averaged higher in amino-acids than those

tempered with the other solutions. Hundredth normal sodium hydroxide, hundredth normal sulfuric acid, tenth normal sodium hydroxide, twentieth normal sodium sulfate, and tenth normal sulfuric acid followed in order of decreasing magnitude in the amino-acid content of wheat when these solutions were used as the tempering agents.

The amino-acids apparently follow Loeb's theory of the chemical behavior of proteins, and combine readily with alkalis upon the alkaline side of neutrality, and with acids upon the acid side of neutrality. The results with sodium sulfate indicate that the amino-acids may combine directly with disassociated salts when the hydrogen ion concentration is close to the iso-electric point of the proteins.

At twenty hours' tempering time, the sample of Hybrid 128 wheat showed a small increase in water soluble nitrogen when tempered with distilled water. It had the same increase with hundredth normal acid and tenth normal acid as with water. Hundredth normal sodium hydroxide gave a much larger increase than water, and tenth normal sodium hydroxide caused double the increase of the hundredth normal sodium hydroxide. The twentieth normal sodium sulfate solution lowered the water soluble nitrogen content.

The water soluble nitrogen content of the sample of Soft Federation wheat tempered with distilled water increased moderately during the first twenty hours, and then increased at a slower rate for the remainder of the

ninety-two hours. All the samples tempered with acid and alkali solutions acted very much alike - each with a greater increase during the first period, and then a decrease at ninety-two hours to about the same value as the water-tempered sample. The sample tempered with twentieth normal sodium sulfate had a smaller increase during the first twenty hours, but was equal to the others at the end of the long period.

The sample of soft red winter wheat increased in water soluble nitrogen when tempered twenty hours with distilled water. After twenty hours there was a falling off to about the original value at ninety-two hours. When tenth normal sulfuric acid was substituted for water, the water soluble nitrogen content was increased about three-tenths of one per-cent during tempering. Tenth normal sodium hydroxide caused a smaller gain than the acid, but there was no decrease in the latter part of the tempering period. Hundredth normal acid and alkali at ninety-two hours caused the water soluble nitrogen content to fall between the water temper and the tenth normal acid temper. The twentieth normal sodium sulfate when used as the tempering agent gave a lesser increase in water soluble nitrogen than did water, but the increase continued during the latter part of the tempering period, giving a final result nearly the same as with tenth normal sulfuric acid.

There was a large increase in water soluble nitrogen content of the hard winter wheat sample when tempered with water, followed by a moderate decrease during the latter part of the tempering period. The sample tempered with tenth normal sodium hydroxide had a larger increase during the first twenty hours, but at ninety-two hours was somewhat lower than the water-tempered sample. The other tempers at twenty hours were grouped a little lower than the water-tempered sample. At ninety-two hours all values were closely grouped except the sample tempered with twentieth normal sodium sulfate solution, which had a much lower water soluble nitrogen content than the others.

As was the case with the amino-acids, the changes in water soluble nitrogen during tempering are functions of the wheat rather than the hydrogen ion concentration of the tempering water. However there seems to be a tendency for the tenth normal acid and alkali to cause a slight increase in the water soluble nitrogen content of a tempered wheat sample.

All the samples except the Hybrid 128 show a small degree of correlation between the titratable acidity and the water soluble nitrogen.

The copper non-precipitable nitrogen content of the Hybrid 128 wheat decreased slightly upon twenty hours' temper with distilled water. When tenth normal acid or

alkali was used instead of water, there was an increase rather than a decrease. Hundredth normal acid and alkali, and twentieth normal sodium sulfate caused greater decreases than did water.

The Soft Federation wheat sample increased in copper non-precipitable nitrogen throughout the entire tempering period when tempered with distilled water. Tenth normal sodium hydroxide gave slightly higher results at twenty hours, but the same as water at ninety-two hours. When tenth normal sulfuric acid was substituted for the water, the copper non-precipitable nitrogen was much higher at twenty hours, but a little lower at ninety-two. Hundredth normal acid and alkali gave somewhat lower results than water. There was a sharp decrease in copper non-precipitable nitrogen when tempered with twentieth normal sodium sulfate at twenty hours. After twenty hours the copper non-precipitable nitrogen content increased to nearly the original value by ninety-two hours.

When the soft red winter wheat was tempered with distilled water, the copper non-precipitable nitrogen first decreased, and then increased at a rapid rate for the remainder of the period of ninety-two hours. The use of tenth normal sodium hydroxide resulted in an increase instead of a decrease. Tenth normal sulfuric acid gave an even larger increase in the first twenty hours.

On the other hand, twentieth normal sodium sulfate caused an even greater decrease than did water. At ninety-two hours the water-tempered sample and the hundredth normal samples showed the same large increase in copper non-precipitable nitrogen. The twentieth normal sodium sulfate sample had nearly as large an increase. The two samples tempered with tenth normal acid and alkali showed much greater increases than the water-tempered sample.

The hard red winter wheat sample showed a large increase in copper non-precipitable nitrogen during the first of the tempering period, and a continued slower increase during the next seventy-two hours. All the acid and alkali tempered samples acted alike. They gave slightly higher results at twenty hours, and then were constant for the rest of the time. The twentieth normal sodium sulfate tempered sample was a little lower than the water-tempered one.

The tenth normal sulfuric acid and the tenth normal sodium hydroxide appear to increase the copper non-precipitable nitrogen when used to temper a sample of wheat. Salt solutions seem to have a decreasing effect. Hundredth normal acid and alkali did not vary enough from the water to give consistently different results.

The effect of varying the hydrogen ion concentration of the tempering water is small. An inspection of the analytical results reveals a tendency for the tenth normal sulfuric acid and the tenth normal sodium hydroxide to increase the decomposition products of the proteins more than is done by tempering with distilled water, the amino-acids being exceptions to this conclusion. Amino-acid content is lowered by the stronger acid or alkali. Hundredth normal acid and alkali are erratic in results. Twentieth normal sodium sulfate tends to retard the breaking down of the proteins during tempering. Sodium hydroxide appears to combine chemically with the wheat more rapidly than does sulfuric acid. Sodium sulfate shows a tendency to combine with the amino-acids.

TABLE 9

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Variable Hydrogen Ion Concentration of the Tempering Water
Hybrid 128 Wheat

Tempered With	Time Temp. Hrs.	Moisture %	Acidity c.c. H ₂ SO ₄	Amino-N %	Water Sol.N %	Cu Non- pp.N %
Dry Wheat	0	9.10	2.4	.0124	.254	.0598
Dist. Water	20	17.37	2.35	.0138	.267	.0578
N/10 H ₂ SO ₄	21	16.45	3.3	.0107	.268	.0650
N/10 NaOH	21	16.12	2.35	.0113	.318	.0626
N/100 H ₂ SO ₄	20	17.30	2.10	.0129	.267	.0519
N/100 NaOH	20	16.73	2.15	.0143	.294	.0545
N/20 Na ₂ SO ₄	18	17.67	1.8	.0130	.250	.0494

Variable Hydrogen Ion Concentration of the Tempering Water
Soft Federation Wheat

Tempered With	Time Temp. Hrs.	Moisture %	Amino-N %	Acidity c.c. H ₂ SO ₄	Water Sol.N %	Cu Non- pp.N %
Dist. Water	0	9.30	.0157	2.4	.290	.0659
" "	21	17.13	.0194	2.25	.306	.0722
" "	92	16.15	.0135	2.55	.314	.0740
N/10 H ₂ SO ₄	20	16.05	.0135	3.35	.325	.0826
" "	92	16.62	.0179	2.85	.317	.0716
N/10 NaOH	20	16.56	.0135	2.25	.320	.0744
" "	92	16.20	.0177	2.05	.320	.0740
N/100 H ₂ SO ₄	20	15.80	.0177	2.7	.324	.0680
" "	92	15.57	.0176	1.9	.302	.0644
N/100 NaOH	20	16.36	.0185	2.65	.321	.0684
" "	92	16.06	.0185	1.95	.313	.0714
N/20 Na ₂ SO ₄	20	16.92	.0150	2.35	.312	.0546
" "	92	16.74	.0172	2.15	.316	.0654

All results except moisture are calculated to 15% moisture basis.

Variable Hydrogen Ion Concentration of the Tempering Water
Soft Red Winter Wheat.

Tempered With	Time Temp. Hrs.	Moisture %	Acidity c.c. H ₂ SO ₄	Amino-N %	Water Sol. N %	Cu Non- pp. N %
Dry Wheat	0	10.63	3.4	.0180	.321	.0880
Dist. Water	20	18.18	3.85	.0204	.346	.0846
" "	92	18.88	3.8	.0198	.325	.1000
N/10 H ₂ SO ₄	20	17.65	4.45	.0203	.377	.0984
" "	92	17.75	4.55	.0188	.344	.1070
N/10 NaOH	20	17.17	3.1	.0221	.358	.0920
" "	92	17.27	3.25	.0201	.361	.1008
N/100 H ₂ SO ₄	92	17.90	3.35	.0210	.328	.0987
N/100 NaOH	92	18.06	3.45	.0203	.343	.1024
N/20 Na ₂ SO ₄	20	17.51	4.1	.0180	.336	.0838
" "	92	17.82	3.9	.0203	.345	.0958

All results except moisture are calculated to 15% moisture basis.

Variable Hydrogen Ion Concentration of the Tempering Water.

Hard Red Winter Wheat

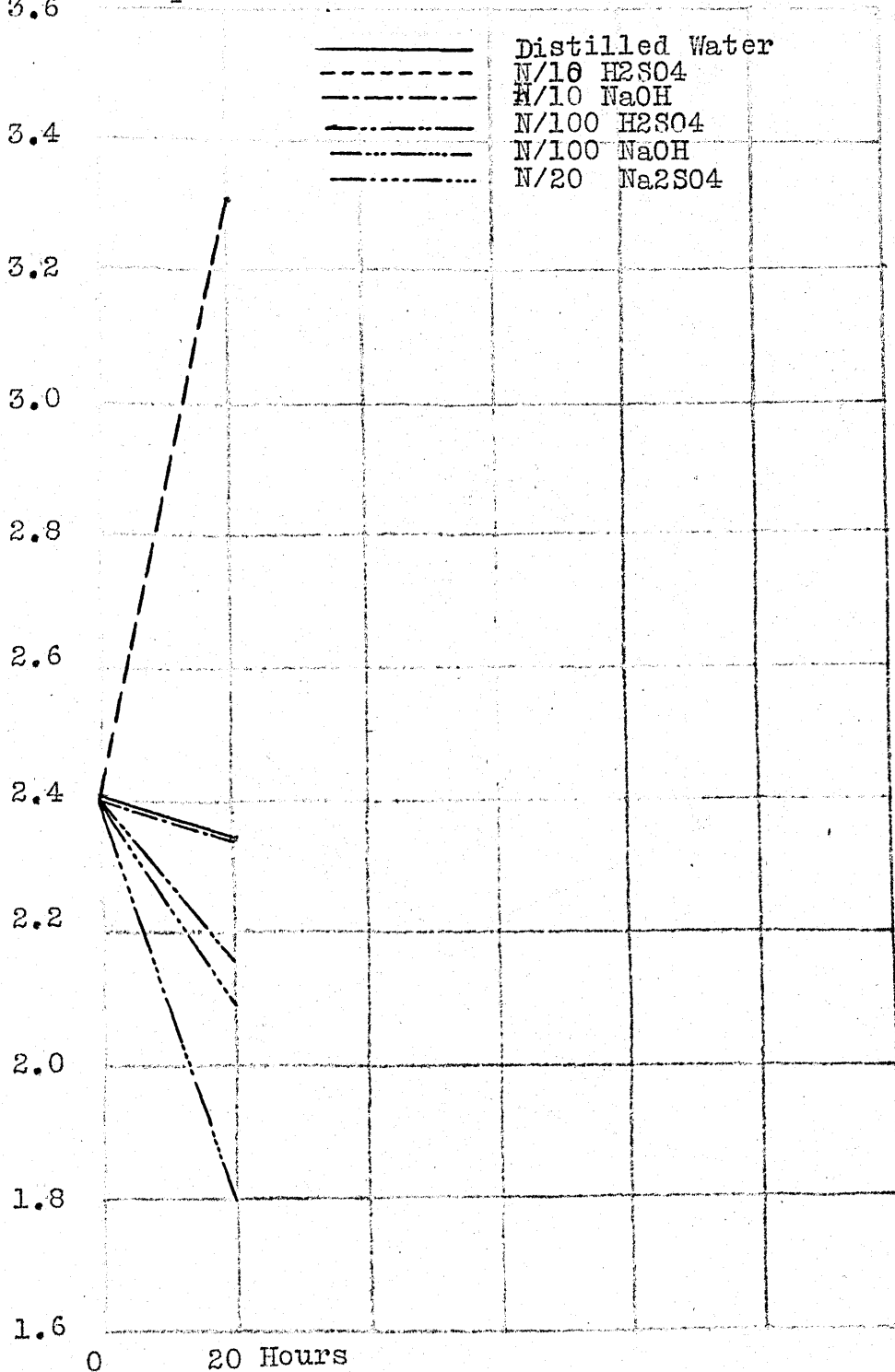
Tempered	Time	Moisture	Acidity	Amino-N	Water	Cu Non-
	Temp.		c.c.		Sol.N	pp.N
With	Hrs.	%	H ₂ SO ₄	%	%	%
Dry Wheat	0	10.78	2.45	.0187	.307	.0696
Distilled Water	20	16.06	3.05	.0213	.341	.0852
" "	92	16.54	2.75	.0164	.333	.0912
N/10 H ₂ SO ₄	20	16.58	3.7	.0171	.332	.0886
" "	92	16.58	3.7	.0178	.322	.0886
N/10 NaOH	20	16.49	2.65	.0164	.356	.0886
" "	92	16.41	2.25	.0143	.327	.0886
N/100 H ₂ SO ₄	20	17.00	2.55	.0179	.338	.0892
" "	92	16.36	2.65	.0185	.319	.0852
N/100 NaOH	20	16.17	3.05	.0156	.331	.0884
" "	92	16.21	2.6	.0171	.322	.0884
N/20 Na ₂ SO ₄	20	16.82	2.6	.0157	.332	.0832
" "	92	15.96	3.55	.0156	.304	.0910

All results except moisture are calculated to 15% moisture basis.

Variable Hydrogen Ion Concentration of Tempering Water

Hybrid 128 Wheat

Titratable Acidity - Tempering Time

c.c. N/10 H_2SO_4
3.6

Variable Hydrogen Ion Concentration of the Tempering Water

Soft Federation Wheat

Titratable Acidity - Tempering Time

c.c.N/10 H₂SO₄

3.8

3.6

3.4

3.2

3.0

2.8

2.6

2.4

2.2

2.0

1.8

Distilled Water

N/10 H₂SO₄

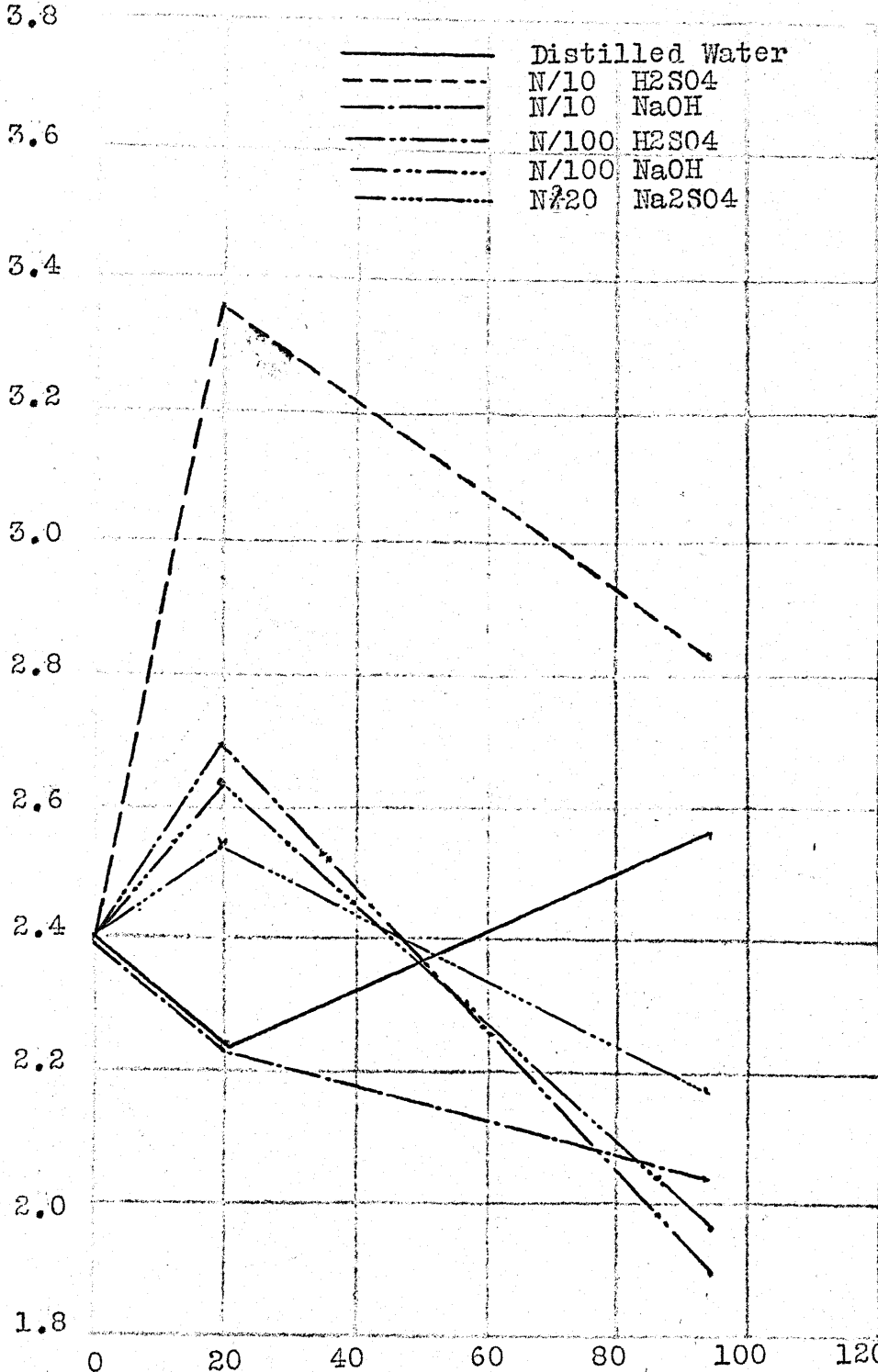
N/10 NaOH

N/100 H₂SO₄

N/100 NaOH

N/20 Na₂SO₄

0 20 40 60 80 100 120 Hours



Variable Hydrogen Ion Concentration of the Tempering Water.

Soft Red Winter Wheat

Titratable Acidity - Tempering Time.

c.c.N/10 H₂SO₄

5.0

4.8

4.6

4.4

4.2

4.0

3.8

3.6

3.4

3.2

3.0

Distilled Water

N/10 H₂SO₄

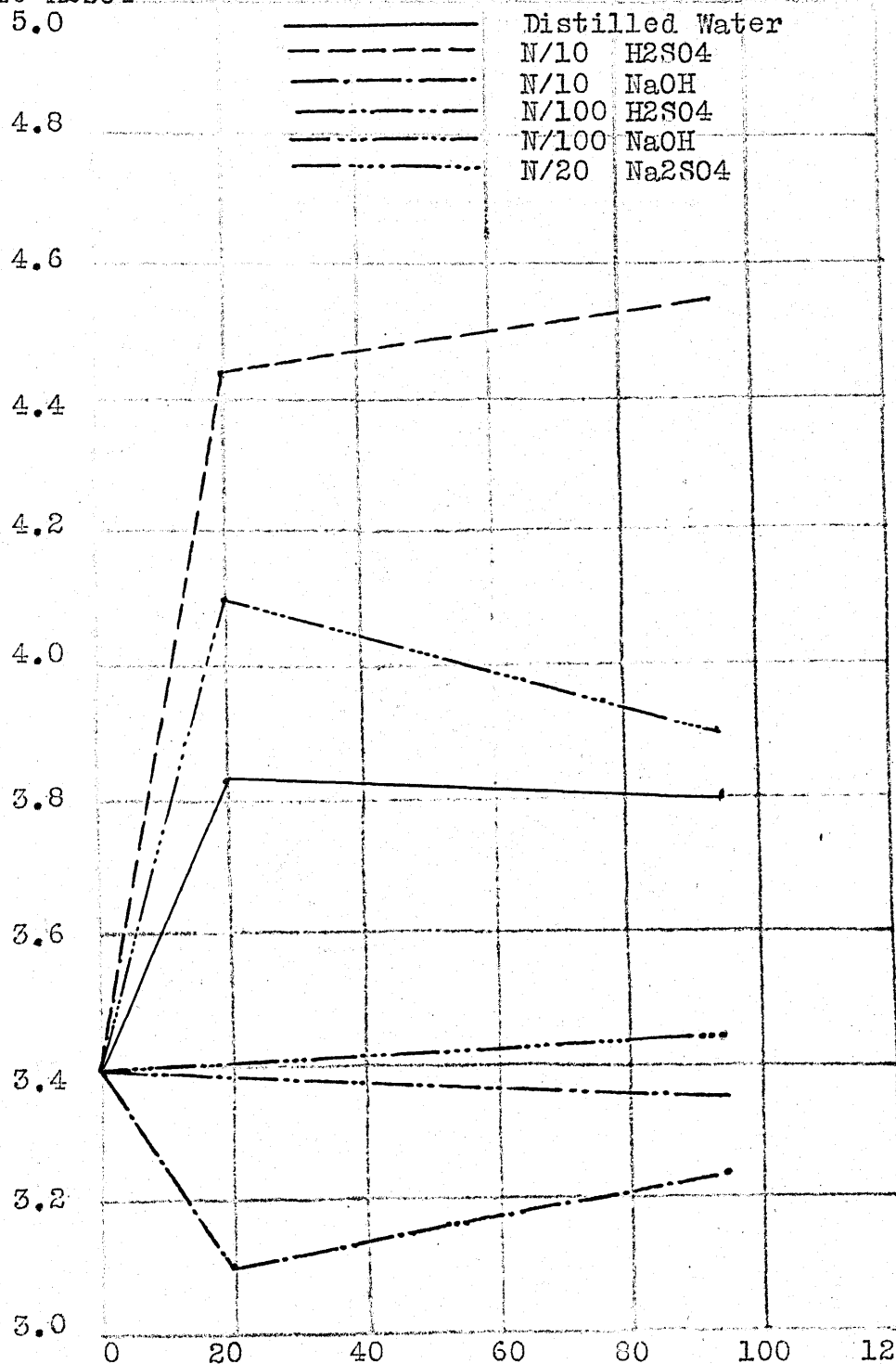
N/10 NaOH

N/100 H₂SO₄

N/100 NaOH

N/20 Na₂SO₄

0 20 40 60 80 100 120 Hours



Variable Hydrogen Ion Concentration of the Tempering Water.

Hard Winter Wheat

Titratable Acidity - Tempering Time

c.c.N/10 H₂SO₄

4.0

3.8

3.6

3.4

3.2

3.0

2.8

2.6

2.4

2.2

2.0

Distilled Water

N/10 H₂SO₄

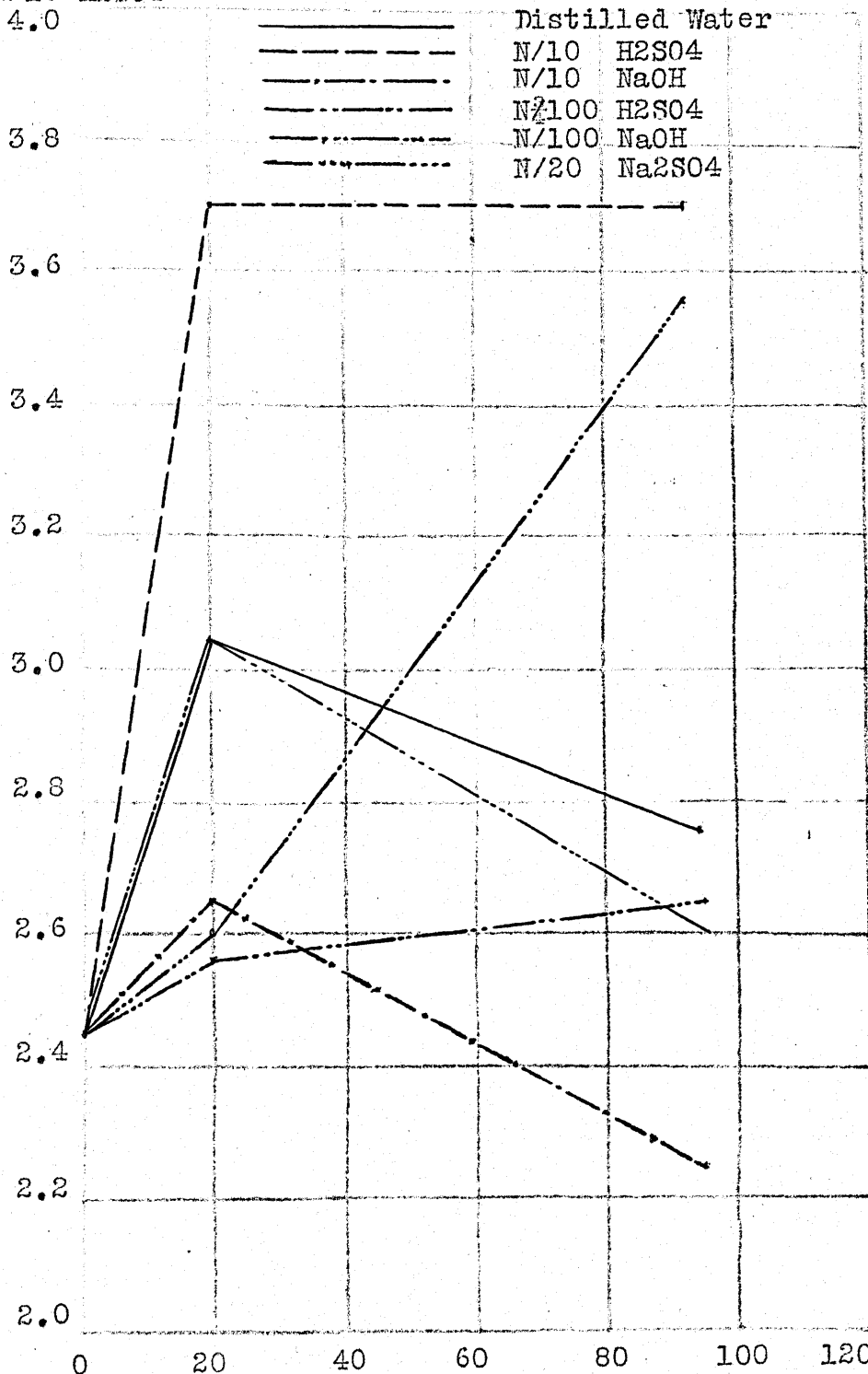
N/10 NaOH

N/100 H₂SO₄

N/100 NaOH

N/20 Na₂SO₄

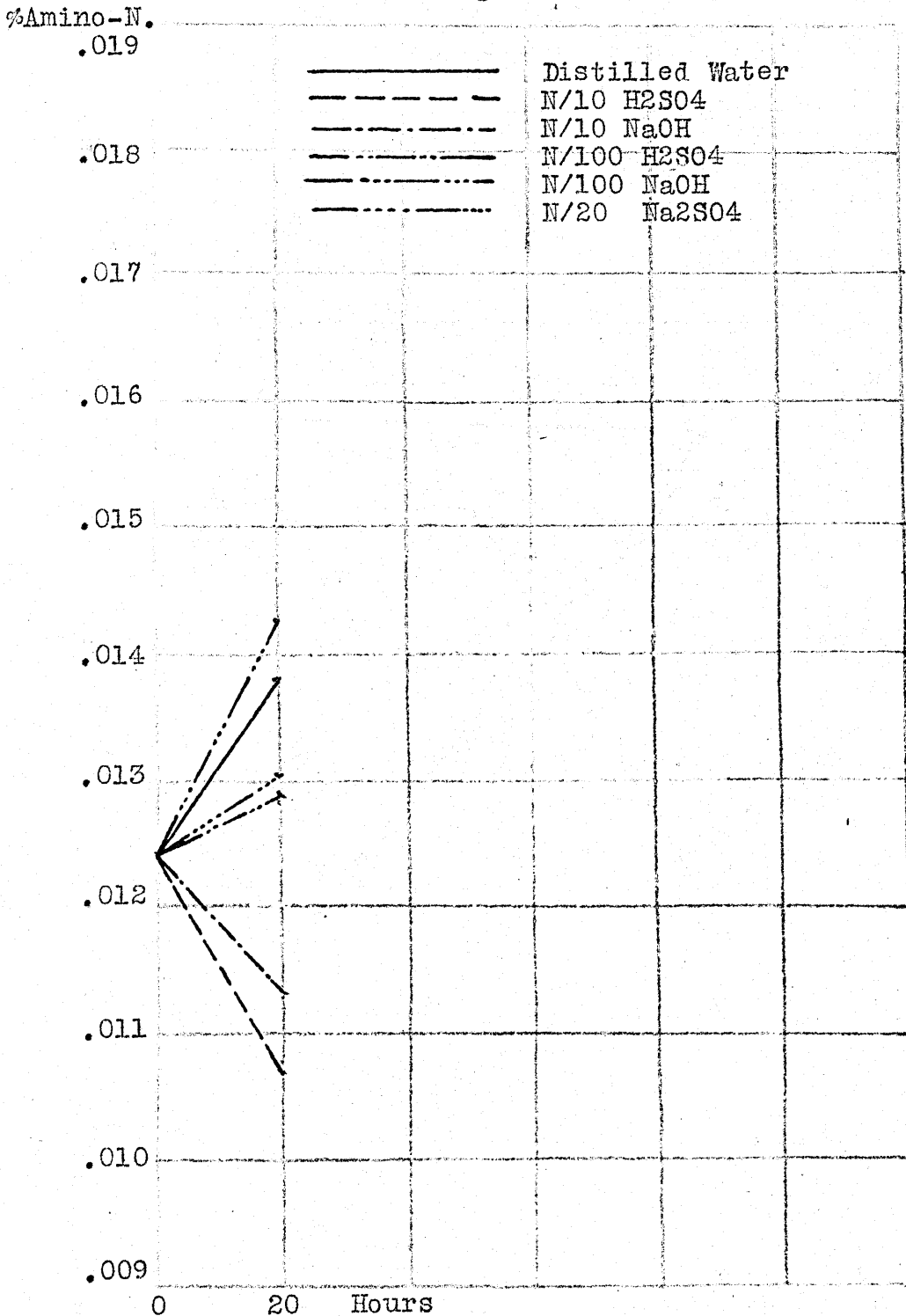
0 20 40 60 80 100 120 Hours



Variable Hydrogen Ion Concentration of the Tempering Water

Hybrid 128 Wheat

Amino-Nitrogen - Tempering Time



Variable Hydrogen Ion Concentration of the Tempering Water.

Soft Federation Wheat.

Amino-Nitrogen - Tempering Time

%Amino-N.

.021

.020

.019

.018

.017

.016

.015

.014

.013

.012

.011

Distilled Water

N/10 H₂SO₄

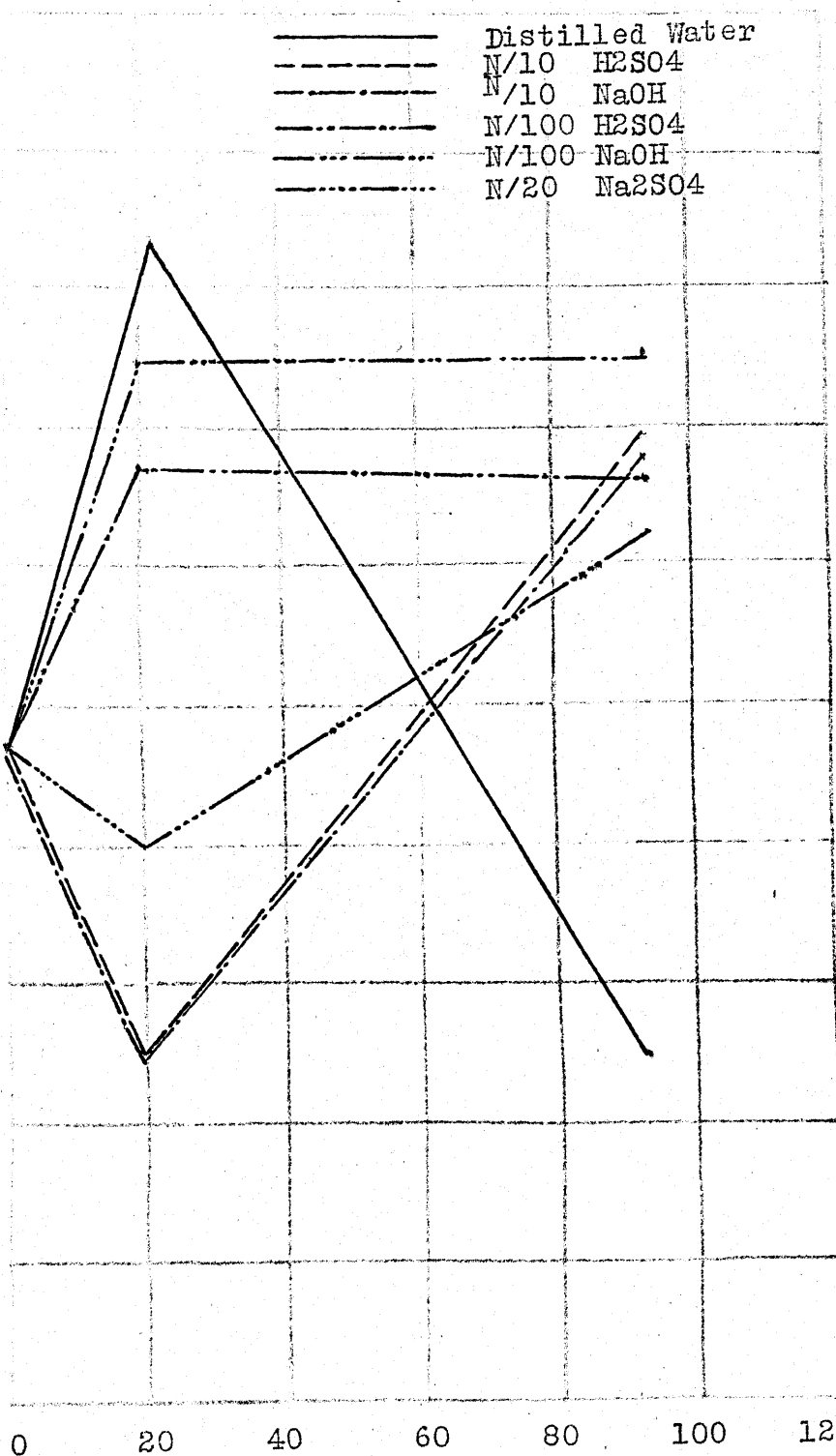
N/10 NaOH

N/100 H₂SO₄

N/100 NaOH

N/20 Na₂SO₄

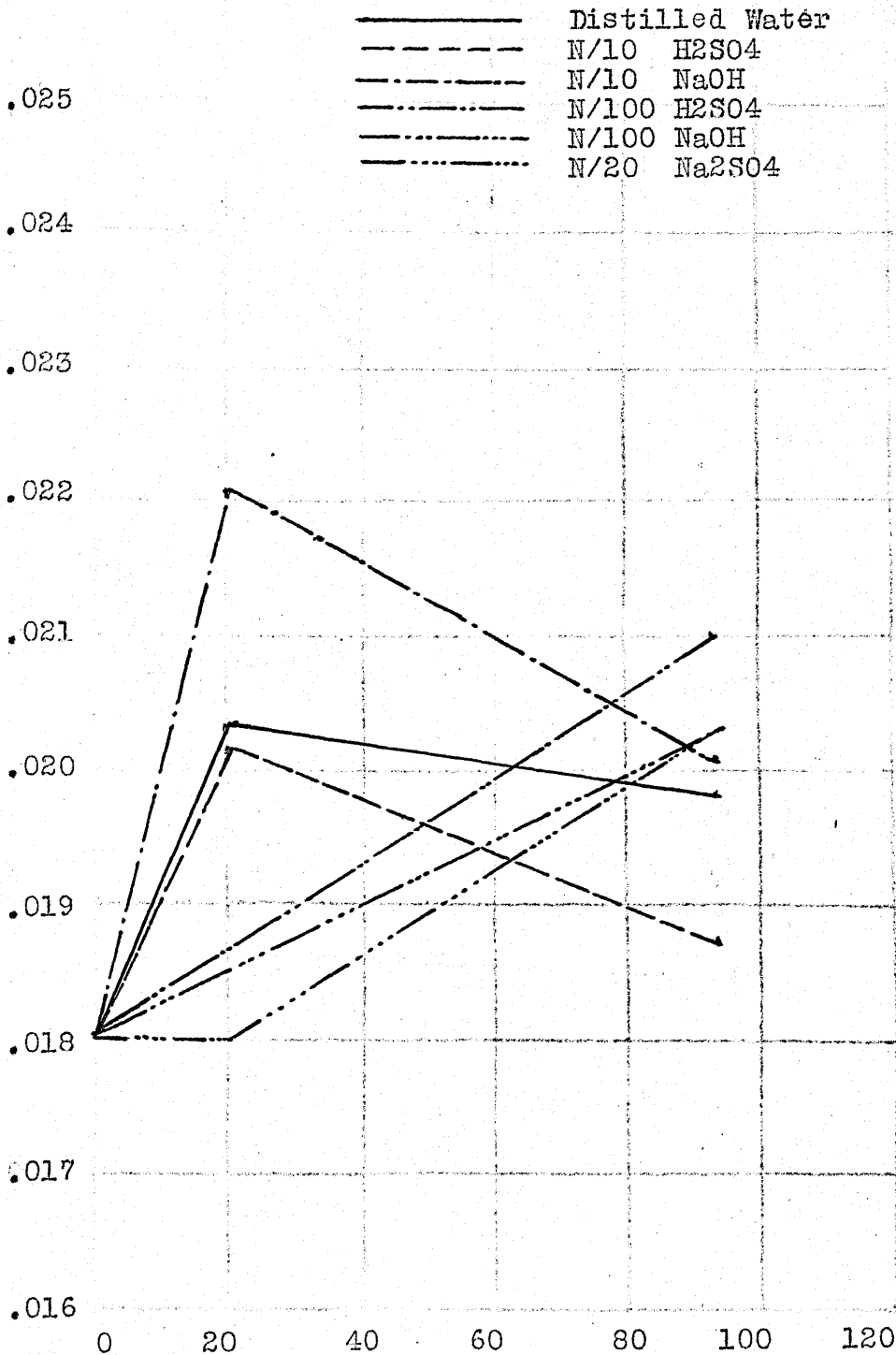
0 20 40 60 80 100 120 Hours



Variable Hydrogen Ion Concentration of the Tempering Water.

Soft Red Winter Wheat

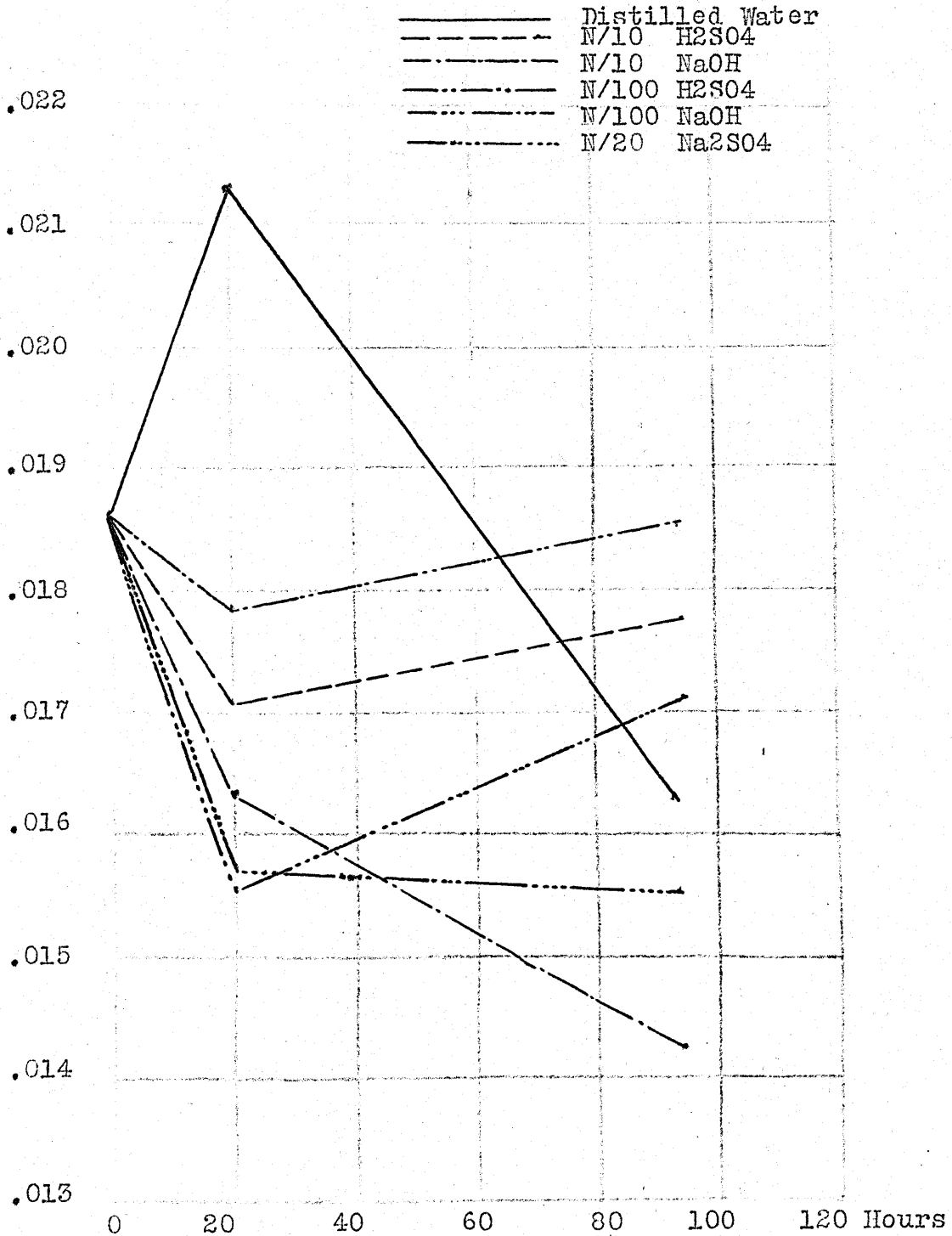
Amino-Nitrogen - Tempering Time

% Amino-N.
.026

Variable Hydrogen Ion Concentration of the Tempering Water

Hard Red Winter Wheat

Amino-Nitrogen - Tempering Time

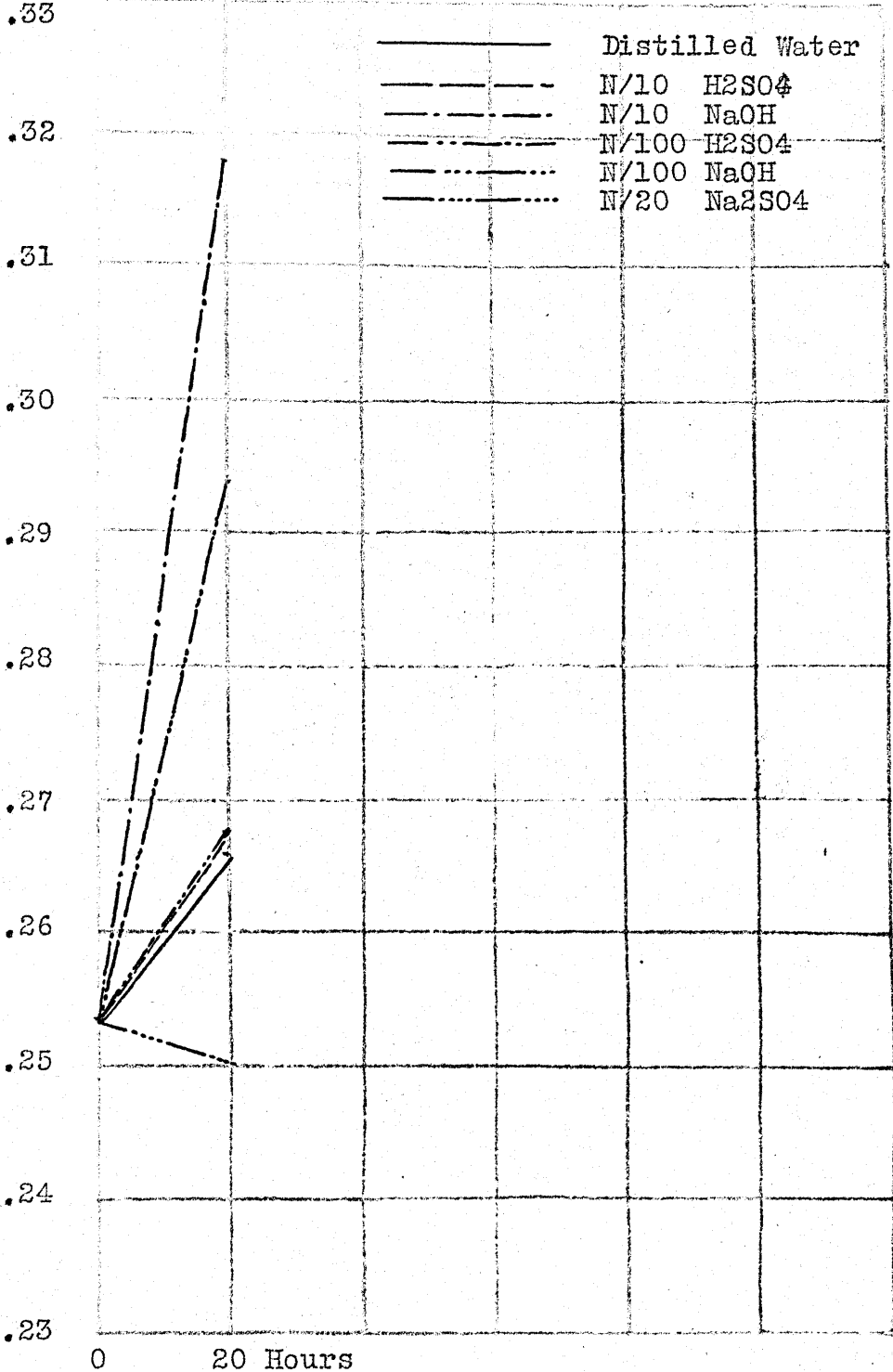
% Amino-N
.023

Variable Hydrogen Ion Concentration of the Tempering Water.

Hybrid 128 Wheat

Water Soluble Nitrogen - Tempering Time

% Water Sol.N.

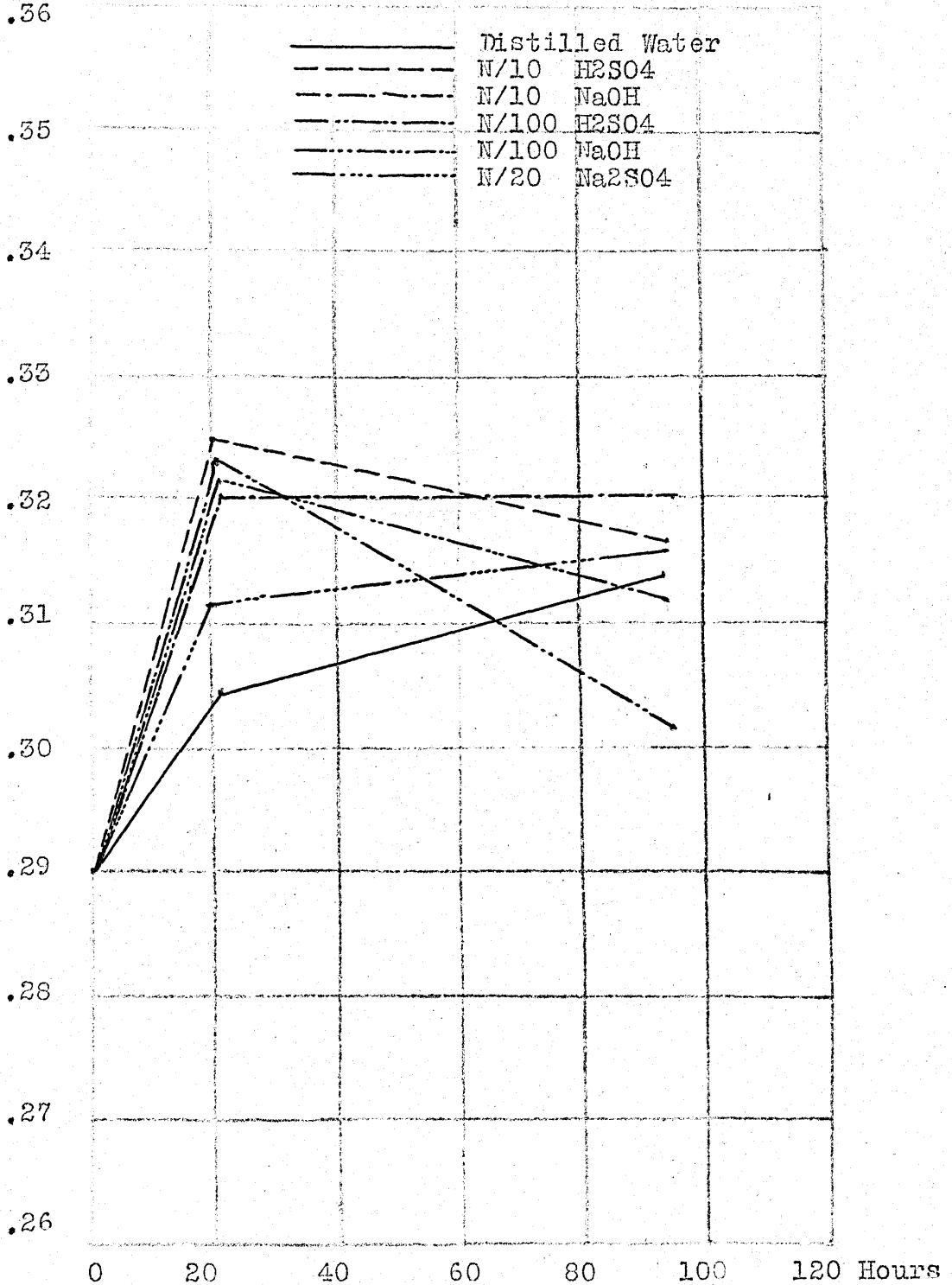


Variable Hydrogen Ion Concentration of the Tempering Water.

Soft Federation Wheat

Water Soluble Nitrogen - Tempering Time

% Water Sol. N.

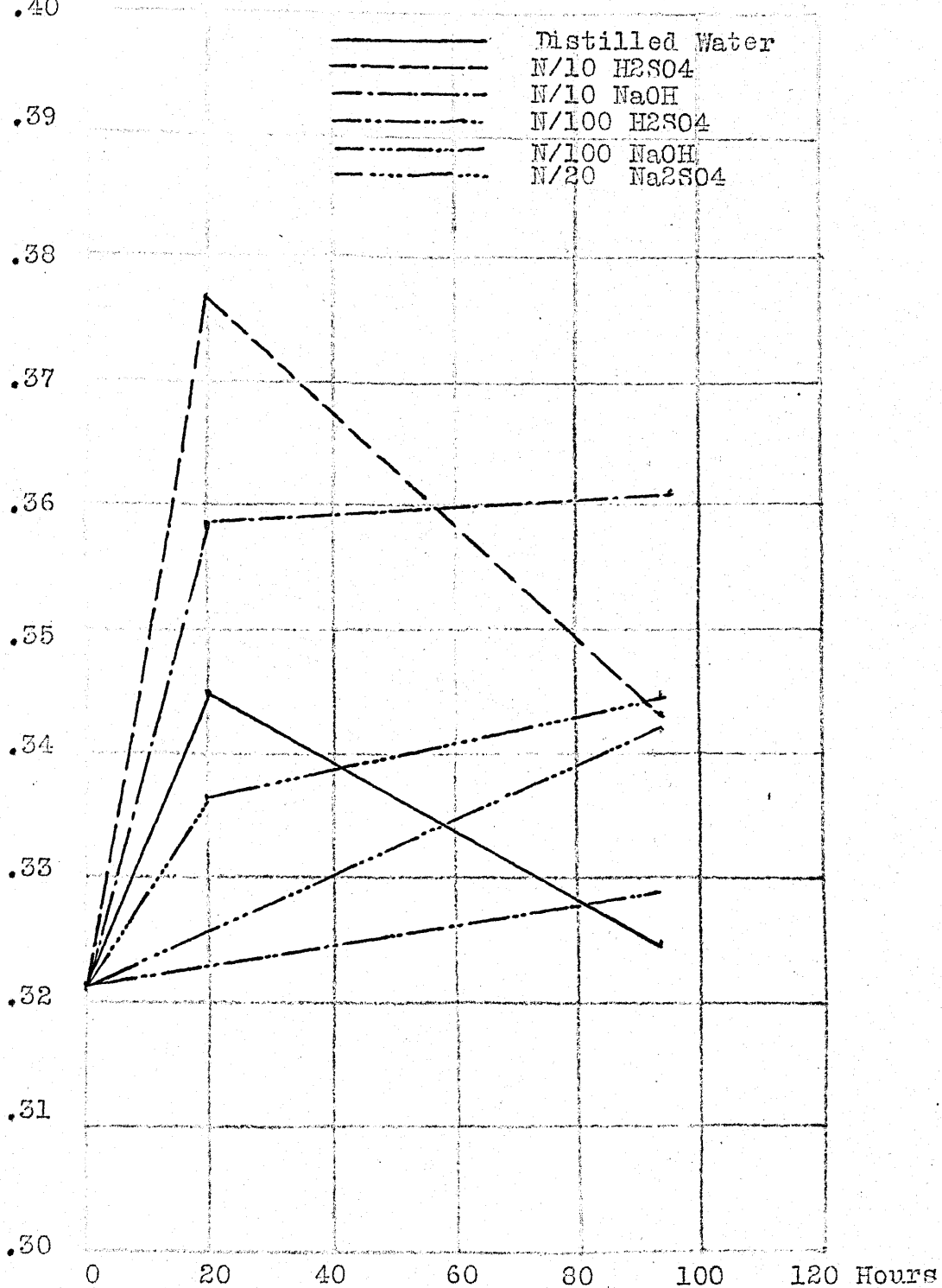


Variable Hydrogen Ion Concentration of the Tempering Water.

Soft Red Winter Wheat

Water Soluble Nitrogen - Tempering Time.

% Water Sol.N.

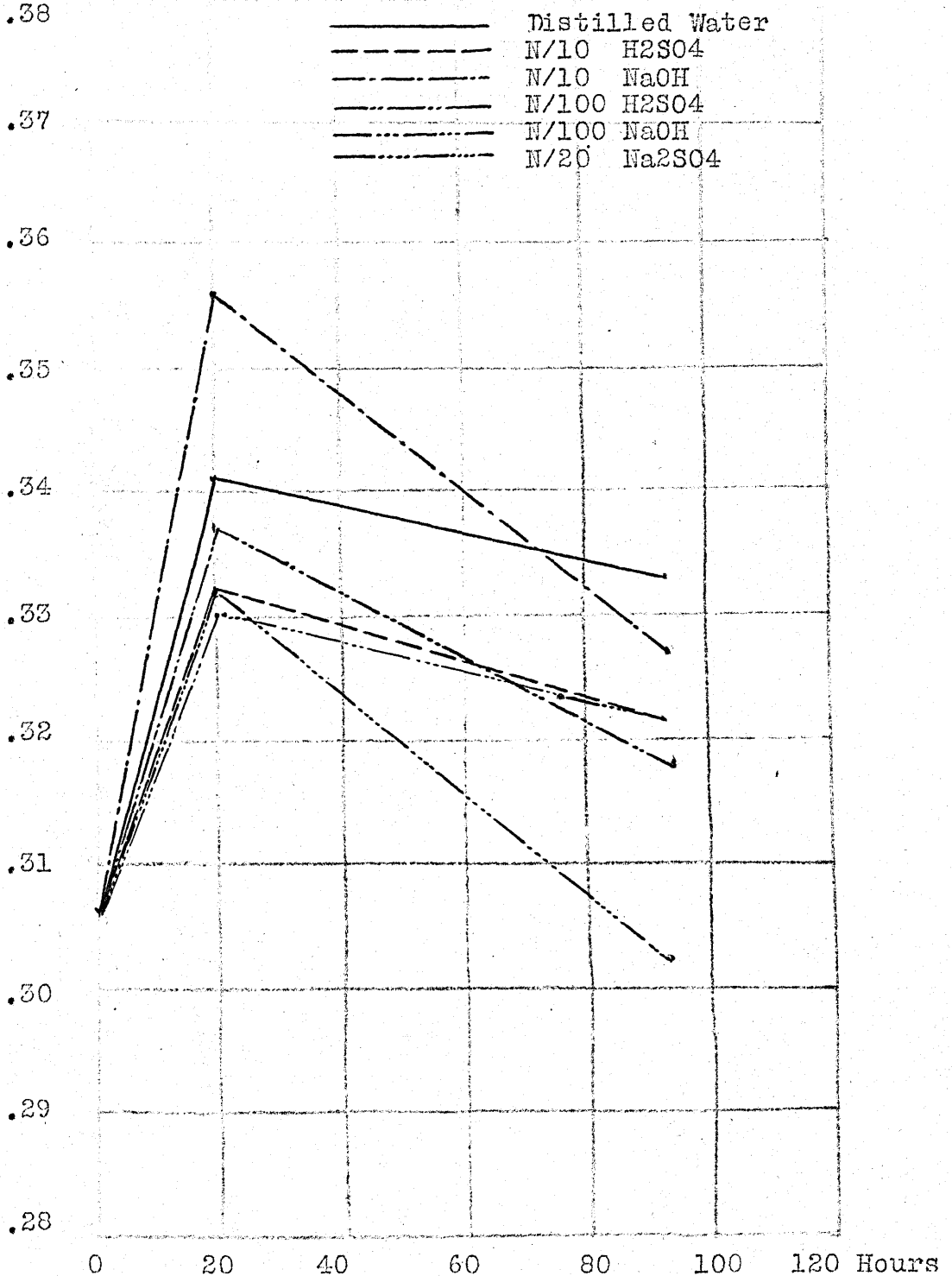


Variable Hydrogen Ion Concentration of the Tempering Water

Hard Red Winter Wheat

Water Soluble Nitrogen - Tempering Water

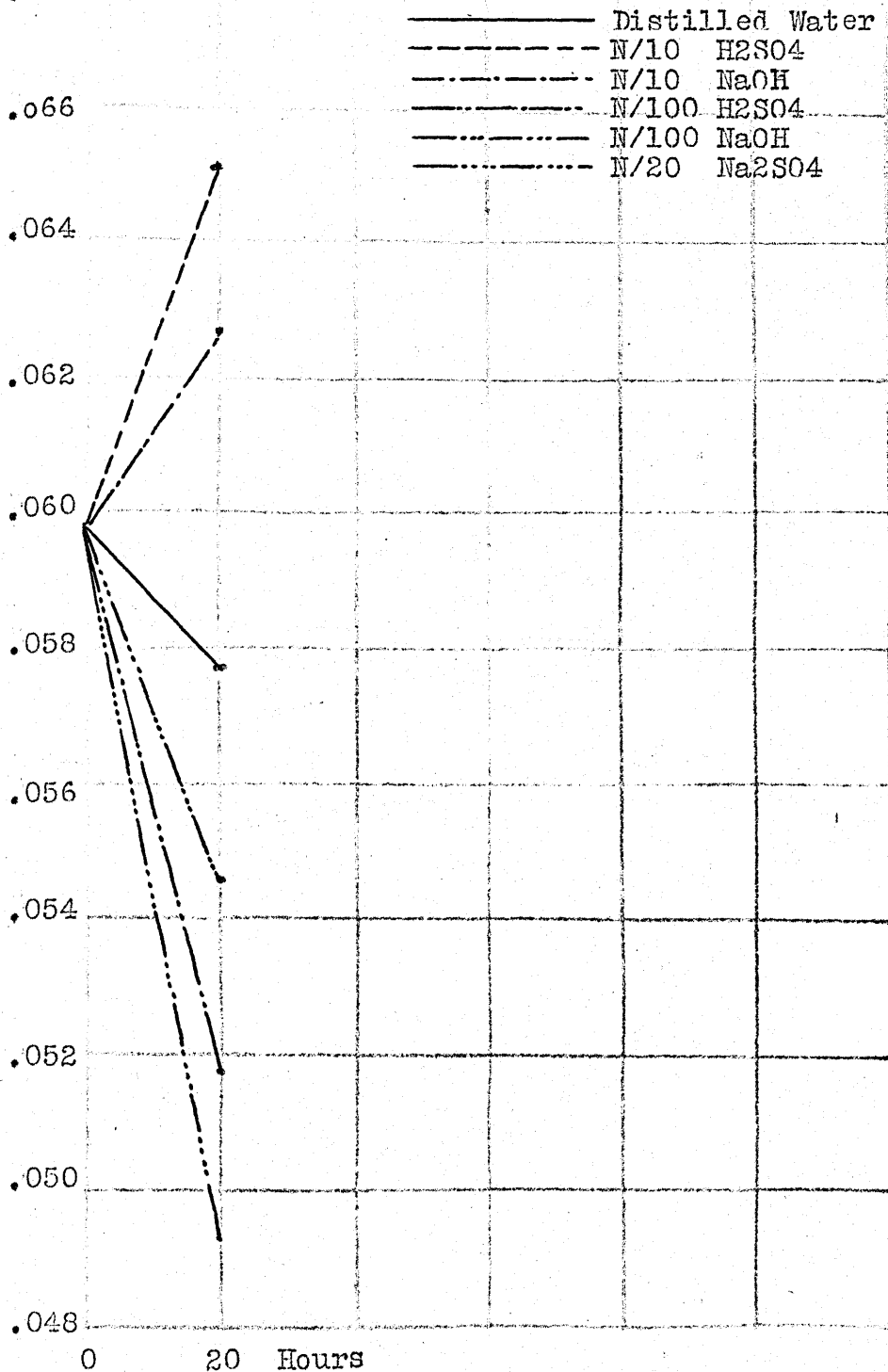
% Water Sol.N.



Variable Hydrogen Ion Concentration of the Tempering Water.

Hybrid 128 Wheat

Copper Non-precipitable. Nitrogen - Tempering Time.

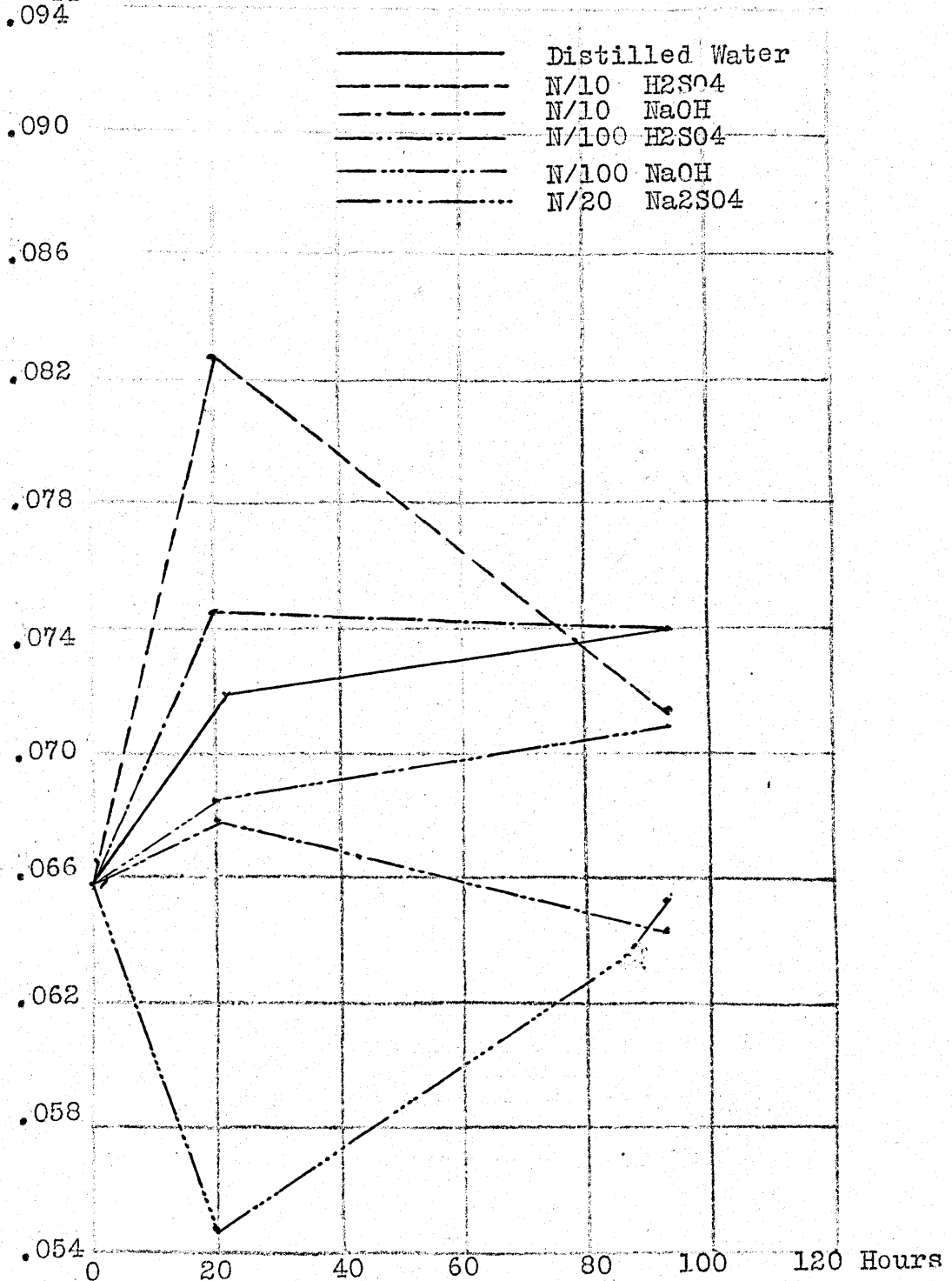
%Cu Non-ppN.
.068

Variable Hydrogen Ion Concentration of the Tempering Water.

Copper Non-precipitable Nitrogen - Tempering Time

Soft Federation Wheat

% Cu non-pp.N.



Variable Hydrogen Ion Concentration of the Tempering Water.

Soft Red Winter Wheat.

Copper Non-precipitable Nitrogen - Tempering Time.

%Cu non-pp.N.

.112

.109

.106

.103

.100

.097

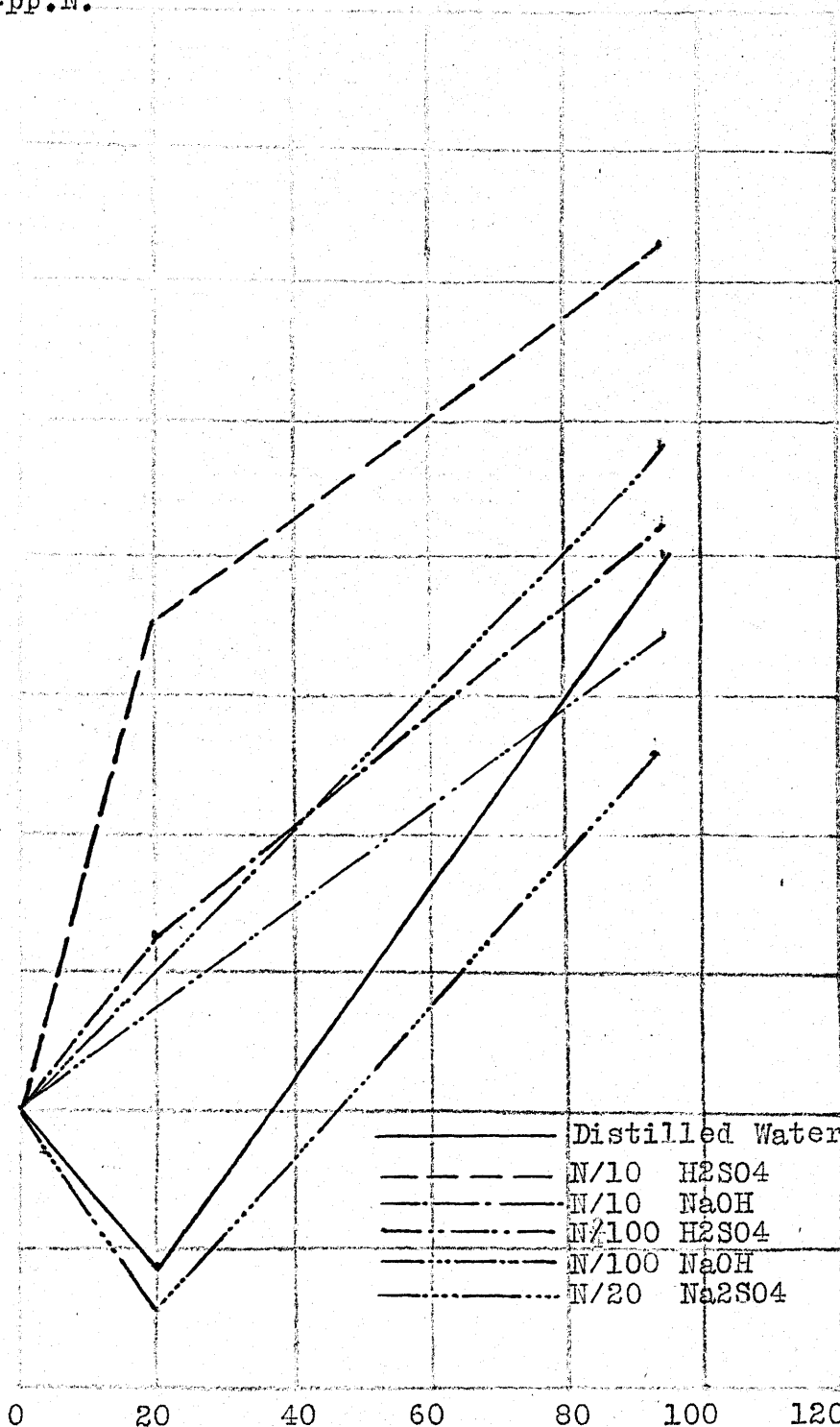
.094

.091

.088

.085

.082



Distilled Water

N/10 H₂SO₄

N/10 NaOH

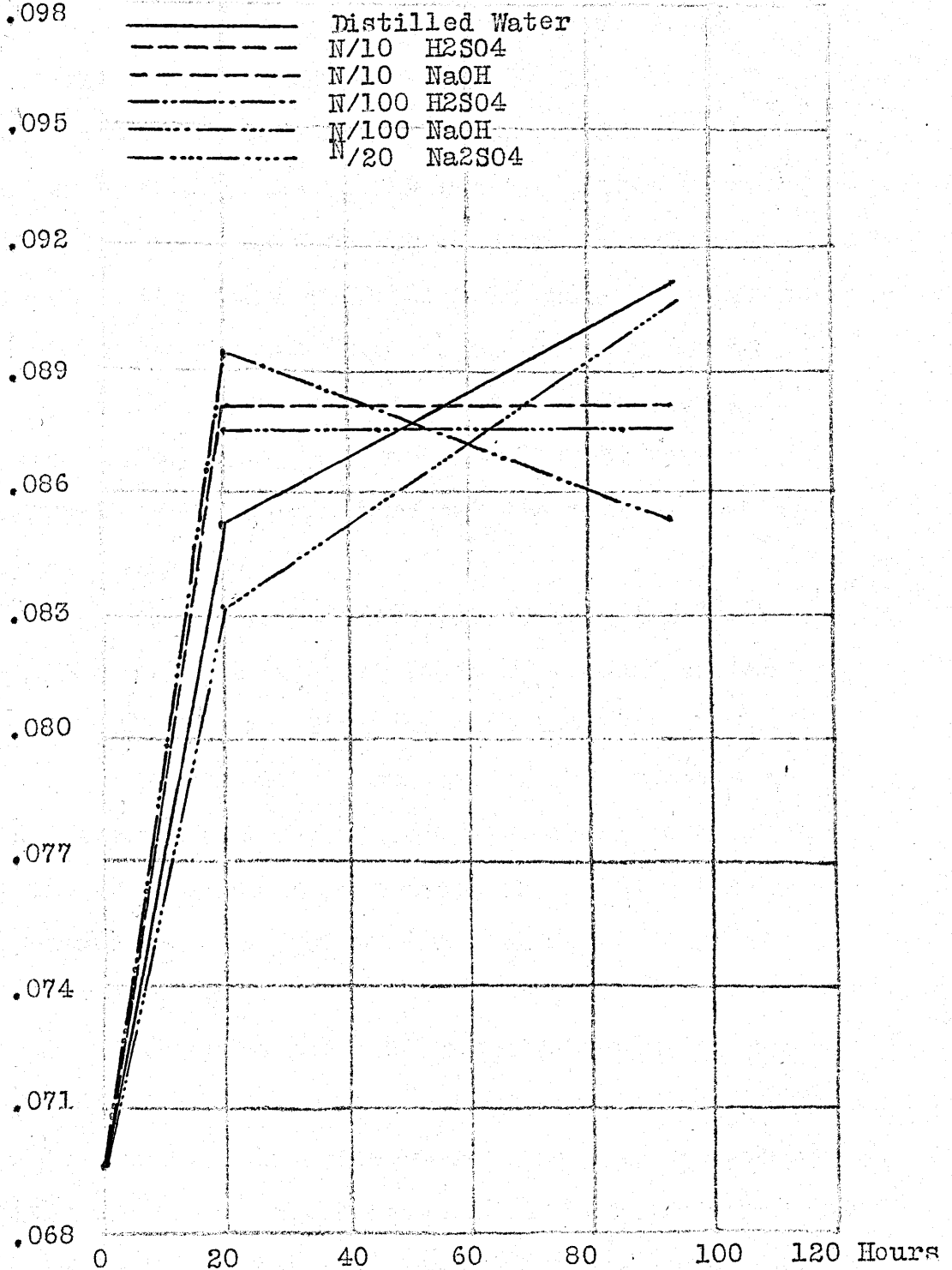
N/100 H₂SO₄

N/100 NaOH

N/20 Na₂SO₄

120 Hours

% Cu non-pp.N.



When water sufficient in quantity to start and maintain germination is added to wheat, the proteins break down into simple forms. When water insufficient in quantity to start germination is added to wheat, desiccation sets in with a building up of the proteins from the simpler nitrogen compounds. When water sufficient in quantity to start germination, but insufficient to maintain it, is added to wheat, the rate of change in the simple nitrogen compounds is a component of the rate of increase during germination and the rate of decrease during desiccation.

The peptide condensation reaction is accelerated both by increasing moisture content of the wheat, and by increasing the temperature.

The chemical behavior of the proteins of any one sample of wheat during tempering is a function of the environmental conditions under which the sample was grown, rather than an inherent function of the variety of wheat.

Tenth normal acid and alkali tend to reduce the amino-acids during the early stages of tempering, and to simultaneously increase the copper non-precipitable nitrogen. The effects of tenth normal acid and alkali upon long tempering are not pronounced. Hundredth

normal acid and alkali have no consistent differences from water, when used to temper wheat. Sodium sulfate solution tends to reduce the quantity of the simpler nitrogenous forms.

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